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=> d hist

     (FILE 'HOME' ENTERED AT 15:32:48 ON 05 NOV 2002)

     FILE 'CAPLUS' ENTERED AT 15:33:28 ON 05 NOV 2002
L1             1 S SULF? (W) (BIND?) (W) GROUP?
L2       1533789 S DISULF? OR SULF? OR THIOL?
L3       2360566 S BIND? OR BOUND? OR TETHER? OR LINK? OR BOND?
L4        139220 S L2 (P) L3
L5       1278804 S DRUG? OR LIGAND? OR TARGET? OR CONJUGATE?
L6         17324 S L4(P) L5
L7       1523034 S INHIBIT?
L8          3861 S L6 (P) L7
L9        241745 S AFFINITY?
L10          845 S L8 (P) L9
L11         6579 S TETHER?
L12            7 S L10 AND L11
L13       343710 S LINK?
L14       349276 S L11 OR L13
L15       120177 S DISULF?
L16         9015 S L14(P)L15
L17        20318 S DRUG(W)DESIGN? OR MOLECULAR(W)RECOGNITION? OR DRUG(W)POTENCY?
L18           10 S L16 (P) L17
L19          132 S L15 (P) L11
L20           17 S L19 (P) L7
L21            6 S L20 (P) L5
L22            7 S L20 (S) L5
L23        36592 S L2 (S) L5
L24           57 S L11 (P) L23
L25           25 S L24 AND PY<=1998
L26            3 S L25 (P) L7
L27            3 S L25 AND L7
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STN SEARCH

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=> d hist

     (FILE 'HOME' ENTERED AT 17:38:07 ON 05 NOV 2002)

     FILE 'CAPLUS' ENTERED AT 17:38:12 ON 05 NOV 2002
L1      1963508 S BIND? OR TETHER? OR LINK? OR COVALENT? OR BOND?
L2      1489213 S CYSTEIN? OR SULF?
L3        35494 S L1 (5N) L2
L4       120177 S DISULF?
L5         2430 S L3 (S) L4
L6       573500 S EXCHANGE?
L7           94 S L5 (P) L6
L8       443780 S LIBRAR? OR COMBINATOR? OR PLURALITY? OR MIXTURE? OR ANALOGS
L9            5 S L7 (P) L8
L10          13 S L5 (S) L8
L11          35 S L5 (S) L6
L12          31 S L11 AND PY<=1998
L13          29 S L11 AND PY<=1997
L14     1523250 S INHIBIT?
L15          35 S L5 (S) L6
L16          35 S L5 (S) L15
L17          75 S L5 (S) L14
L18           5 S L17 (P) AFFINITY
L19           5 S L18 AND PY<=1998
L20        1870 S RSSH OR SULFUR(W)SULFUR
L21        1451 S L1(S) L20
L22          29 S L21 (S) L14
L23          27 S L22 AND PY<=1998
L24           2 S L23 AND L8
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show files
File 155:MEDLINE(R) 1966-2002/Jul W1
File   5:Biosis Previews(R) 1969-2002/Jun W5
         (c) 2002 BIOSIS
File 315:ChemEng & Biotec Abs 1970-2001/Dec
         (c) 2002 DECHEMA
File  73:EMBASE 1974-2002/Jun W5
         (c) 2002 Elsevier Science B.V.
File 399:CA SEARCH(R) 1967-2002/UD=13627
         (c) 2002 AMERICAN CHEMICAL SOCIETY
File 351:Derwent WPI 1963-2002/UD,UM &UP=200242
         (c) 2002  Thomson Derwent
?ds

Set     Items   Description
S1      12031   FLUOROPHORE? ?
S2     172063   LABEL? ?
S3      84573   REPORTER? ?
S4    5947691   AFTER
S5     698324   POST
S6     896514   SUBSEQUENT?
S7      12135   (S1-S3) (10N) (S4-S6)
S8        505   S7 AND (LYSINE? ? OR CYSTEINE? ? OR CARBOXYLIC? ? OR GLUTA-
              MIC OR ASPARTIC)
S9          3   S8 AND (POST()LABELING)
S10         7   S8 AND (SUBSEQUENTLY(5N) (LABELED OR LABELLED))
S11        10   S9 OR S10
S12         6   RD S11 (unique items)
S13        94   S7 (5N) (LYSINE? ? OR CYSTEIN? ? OR CARBOXYLIC? ? OR GLUTA-
              MIC OR ASPARTIC)
S14         7   S13 AND (COUPL? OR LINK? OR CROSSLINK?)
S15         5   RD S14 (unique items)
S16        11   S12 OR S15
S17       498   S8 NOT S14
S18       173   S17 AND (ASSAY? OR DETECT? OR MEASUR?)
S19        11   S18 AND (SPECIFIC(5N) (LYSINE? ? OR CYSTEINE? ? OR CARBOXY-
              LIC OR GLUTAMIC OR ASPARTIC))
S20         7   RD S19 (unique items)
S21        16   S16 OR S20
S22       487   S17 NOT S19
S23       913   POST()LABEL?
S24        18   S23 (5N) (PROTEIN? ? OR POLYPEPTIDE? ?)
S25        12   RD S24 (unique items)
S26        28   S21 OR S25
S27     10349   (SPECIFIC(5N) (LYSINE? ? OR CYSTEINE? ? OR CARBOXYLIC OR G-
              LUTAMIC OR ASPARTIC))
S28        48   S27 (5N) (S1-S3)
S29        27   RD S28 (unique items)
S30        53   S26 OR S29
S31        18   S28 AND (ASSAY? OR DETECT? OR MEASUR?)
S32        10   RD S31 (unique items)
S33        54   S30 OR S32
?t 33/7/all

 33/7/1     (Item 1 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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13319176 22086203 PMID: 11983705
Constitutive Activation of Angiotensin II Type 1 Receptor Alters the Orientation of Transmembrane Helix-2.
Miura Shin-Ichiro; Karnik Sadashiva S
Department of Molecular Cardiology, Lerner Research Institute, The Cleveland Clinic Foundation, Cleveland, Ohio 44195.
Journal of biological chemistry (United States) Jul 5 2002, 277 (27) p24299-305, ISSN 0021-9258 Journal Code: 2985121R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: In Process
A key step in transmembrane (TM) signal transduction by G-protein-coupled receptors (GPCRs) is the ligand-induced conformational change of the receptor, which triggers the activation of a guanine nucleotide-binding protein. GPCRs contain a seven-TM helical structure essential for signal transduction in response to a large variety of sensory and hormonal signals. Primary structure comparison of GPCRs has shown that the second TM helix contains a highly conserved Asp residue, which is critical for agonist activation in these receptors. How conformational changes in TM2 relate to signal transduction by a GPCR is not known, because activation-induced conformational changes in TM2 helix have not been measured . Here we use modification of reporter cysteines to measure water accessibility at specific residues in TM2 of the type 1 receptor for the octapeptide hormone angiotensin II. Activation-dependent changes in the accessibility of Cys(76) on TM2 were measured in constitutively activated mutants. These changes were directly correlated with measurement of function, establishing the link between physical changes in TM2 and function. Accessibility changes were measured at several consecutive residues on TM2, which suggest that TM2 undergoes a transmembrane movement in response to activation. This is the first report of in situ measurement of TM2 movement in a GPCR.
Record Date Created: 20020701

33/7/2 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

13316980 22047854 PMID: 12051849
Knowledge-based design of reagentless fluorescent biosensors from recombinant antibodies.
Renard Martial; Belkadi Laurent; Hugo Nicolas; England Patrick; Altschuh Daniele; Bedouelle Hugues
Departement de Biologie Structurale et Chimie, CNRS URA 2185, Institut Pasteur, 28 rue Docteur Roux, 75724 Paris Cedex 15, France.
Journal of molecular biology (England) Apr 26 2002, 318 (2) p429-42, ISSN 0022-2836 Journal Code: 2985088R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The possibility of obtaining from any antibody a fluorescent conjugate which responds to the binding of the antigen by a variation of its fluorescence, would be of great interest in the analytical sciences and for the construction of protein chips. This possibility was explored with antibody mAbD1.3 directed against hen egg white lysozyme. Rules of design were developed to identify the residues of the antibody to which a

fluorophore could be chemically coupled , after changing them to cysteine by mutagenesis. These rules were based on: the target residue belonging to a topological neighbourhood of the antigen in the structure of the complex between antibody and antigen; its absence of functional importance for the interaction with the antigen; and its solvent accessibility in the structure of the free antibody. Seventeen conjugates between the single-chain variable fragment scFv of mAbD1.3 and an environment-sensitive fluorophore were constructed. For six of the ten residues which fully satisfied the design rules, the relative variation of the fluorescence intensity between the free and bound states of the conjugate was comprised between 12 and 75% (in non-optimal buffer), and the affinity of the conjugate for lysozyme remained unchanged relative to the parental scFv. In contrast, such results were true for only one of the seven residues which failed to satisfy one of the rules and were used as controls. One of the conjugates was studied in more detail. Its fluorescence increased proportionally to the concentration of lysozyme in a nanomolar range, up to 90% in a defined buffer, and 40% in serum. This increase was specific for hen egg lysozyme and it was not observed with a closely related protein, turkey egg lysozyme. The residues which gave operational conjugates (six in V(L) and one in V(H)), were located in the immediate vicinity of residues which are functionally important, along the sequence of FvD1.3. The results suggest rules of design for constructing antigen-sensitive fluorescent conjugates from any antibody, in the absence of structural data. (c) 2002 Elsevier Science Ltd.
Record Date Created: 20020607

33/7/3 (Item 3 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

12895804 21476689 PMID: 11592693
EPR studies of iso-1-cytochrome c: effect of temperature on two-component spectra of spin label attached to cysteine at positions 102 and 47.
Pyka J; Osyczka A; Turyna B; Blicharski W; Froncisz W
Institute of Molecular Biology, Jagiellonian University, Krakow, Poland.
European biophysics journal : EBJ (Germany) Sep 2001, 30 (5) p367-73 , ISSN 0175-7571 Journal Code: 8409413
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Wild-type iso-1-cytochrome c from Saccharomyces cerevisiae containing naturally occurring cysteine at position 102 and mutated protein S47C (derived from the protein in which C102 had been replaced by threonine) were labeled with cysteine - specific methanethiosulfonate spin label . Continuous wave (CW) electron paramagnetic resonance (EPR) was used to examine the effect of temperature on the behavior of the spin label in the oxidized and reduced forms of wild-type cytochrome c and in the oxidized form of the mutated protein. The computer simulations revealed that the CW EPR spectrum for each form of cytochrome c consists of at least two components [a fast (F) and a slow (S) component], which differ in the values of the rotational correlation times tauRparallel (longitudinal rotational correlation time) and tauRperpendicular (transverse rotational correlation time) and that the relative contributions of the F and S components of the spectra change with temperature. In addition, the values of the rotational correlation times (tauRparallel and tauRperpendicular) for the F component appear to change much more dramatically with the

temperature than the respective values for the S component. A large difference between the behavior of the oxidized and reduced wild-type spin-labeled cytochromes c indicates that the temperature-induced unfolding of the protein in the region around C102 progresses more rapidly when cytochrome c is in the oxidized form.
Record Date Created: 20011010

33/7/4 (Item 4 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

12520300 21358643 PMID: 11464508
Detection of fluorescence dye-labeled proteins in 2-D gels using an Arthur 1442 Multiwavelength Fluoroimager.
Herick K; Jackson P; Wersch G; Burkovski A
PerkinElmer Life Sciences/Wallac Oy, Turku, Finland. klaus.herick@perkinelmer.com
BioTechniques (United States) Jul 2001, 31 (1) p146-9, ISSN 0736-6205 Journal Code: 8306785
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Labeling of proteins with SYPRO Orange, SYPRO Red, and SYPRO Ruby after 2-D polyacrylamide gel electrophoresis (PAGE) using plastic-backed immobilized pH gradient (IPG) strips and precast SDS polyacrylamide gels was tested. Protein spots were detected using an Arthur 1442 Multiwavelength Fluoroimager. The labeling methods described allow detection of proteins both after isoelectric focusing (IEF) and PAGE with a sensitivity higher than or comparable to standard silver staining methods. In addition to the post - labeling methods mentioned above, pre-labeling with the cysteine - specific fluorophore monobromobimane before 2-D PAGE is a sensitive, fast, and cost-effective alternative to existing staining protocols.
Record Date Created: 20010723

33/7/5 (Item 5 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

11355726 21429493 PMID: 11544597
Screening for disulfide-rich peptides in biological sources by carboxyamidomethylation in combination with differential matrix-assisted laser desorption/ionization time-of-flight mass spectrometry.
Neitz S; Jurgens M; Kellmann M; Schulz-Knappe P; Schrader M
BioVisioN GmbH & Co. KG, Feodor-Lynen-Str. 5, 30625 Hannover, Germany.
Rapid communications in mass spectrometry : RCM (England) 2001, 15 (17) p1586-92, ISSN 0951-4198 Journal Code: 8802365
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Peptides with biological functions often contain disulfide bridges connecting two cysteine residues. In an attempt to screen biological fluids for peptides containing cysteine residues, we have developed a sensitive and specific method to label cysteines selectively and detect the resulting molecular mass shift by differential mass spectrometry. First,

reduction of disulfide bridges and carboxyamidomethylation of free thiols is adjusted to quantitatively achieve cysteine alkylation for complex peptide extracts. In a second step, matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOFMS) before and after chemical derivatization is performed, followed by differential analysis to determine shifted peaks; shifted peaks belong to cysteine-containing peptides, other peaks remain unchanged. The number of cysteines can then be determined by the resulting molecular mass shift. Free, reduced cysteines are shifted by 57 u, two oxidized cysteines involved in disulfide bridges (cystine) result in a shift to higher mass per disulfide bridge of 116 u. Disulfide bridges connecting different amino acid chains like insulin break up during reduction. In this case, two peaks with lower molecular masses result from a single one in the unmodified sample. With this technique, we were able to identify cysteine-containing peptides and short fragments of proteins present in human blood filtrate.

Record Date Created: 20010906

33/7/6 (Item 6 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

11352916 21423966 PMID: 11435433
Electron spin resonance and fluorescence studies of the bound-state conformation of a model protein substrate to the chaperone SecB.
Panse V G; Beena K; Philipp R; Trommer W E; Vogel P D; Varadarajan R
Molecular Biophysics Unit, Indian Institute of Science, Bangalore 560 012, India.
Journal of biological chemistry (United States) Sep 7 2001, 276 (36) p33681-8, ISSN 0021-9258 Journal Code: 2985121R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
SecB is a homotetrameric, cytosolic chaperone that forms part of the protein translocation machinery in Escherichia coli. We have investigated the bound-state conformation of a model protein substrate of SecB, bovine pancreatic trypsin inhibitor (BPTI) as well as the conformation of SecB itself by using proximity relationships based on site-directed spin-labeling and pyrene fluorescence methods. BPTI is a 58-residue protein and contains three disulfide groups between residues 5 and 55, 14 and 38, as well as 30 and 51. Mutants of BPTI that contained only a single disulfide were reduced, and the free cysteines were labeled with either thiol- specific spin labels or pyrene maleimide. The relative proximity of the labeled residues was studied using either electron spin resonance spectroscopy or fluorescence spectroscopy. The data suggest that SecB binds a collapsed coil of reduced unfolded BPTI, which then undergoes a structural rearrangement to a more extended state upon binding to SecB. Binding occurs at multiple sites on the substrate, and the binding site on each SecB monomer accommodates less than 21 substrate residues. In addition, we have labeled four solvent-accessible cysteine residues in the SecB tetramer and have investigated their relative spatial arrangement in the presence and absence of the substrate protein. The electron spin resonance data suggest that these cysteine residues are in close proximity (15 A) when no substrate protein is bound but move away to a distance of greater than 20 A when SecB binds substrate. This is the first direct evidence of a conformational change in SecB upon binding of a substrate

protein.
Record Date Created: 20010904

33/7/7 (Item 7 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

10748489 20282971 PMID: 10824857
Probing iso-1-cytochrome c structure by site-directed spin labeling and electron paramagnetic resonance techniques.
Pyka J; Osyczka A; Turyna B; Blicharski W; Froncisz W
Department of Biophysics, Institute of Molecular Biology, Jagiellonian University, Krakow, Poland.
Acta biochimica Polonica (POLAND) 1999, 46 (4) p889-99, ISSN 0001-527X Journal Code: 14520300R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A cysteine - specific methanethiosulfonate spin label was introduced into yeast iso-1-cytochrome c at three different positions. The modified forms of cytochrome c included: the wild-type protein labeled at naturally occurring C102, and two mutated proteins, S47C and L85C, labeled at positions 47 and 85, respectively (both S47C and L85C derived from the protein in which C102 had been replaced by threonine). All three spin-labeled protein derivatives were characterized using electron paramagnetic resonance (EPR) techniques. The continuous wave (CW) EPR spectrum of spin label attached to L85C differed from those recorded for spin label attached to C102 or S47C, indicating that spin label at position 85 was more immobilized and exhibited more complex tumbling than spin label at two other positions. The temperature dependence of the CW EPR spectra and CW EPR power saturation revealed further differences of spin-labeled L85C. The results were discussed in terms of application of the site-directed spin labeling technique in probing the local dynamic structure of iso-1-cytochrome c.
Record Date Created: 20000626

33/7/8 (Item 8 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

10260810 99249871 PMID: 10233084
Azide reduces the hydrophobic barrier of the bacteriorhodopsin proton channel.
Steinhoff H J; Pfeiffer M; Rink T; Burlon O; Kurz M; Riesle J; Heuberger E; Gerwert K; Oesterhelt D
Lehrstuhl fur Biophysik, Ruhr-Universitat Bochum, 44780 Bochum, Germany. hjs@bph.ruhr-uni-bochum.de
Biophysical journal (UNITED STATES) May 1999, 76 (5) p2702-10, ISSN 0006-3495 Journal Code: 0370626
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The sensitivity of a nitroxide spin label to the polarity of its environment has been used to estimate the hydrophobic barrier of the proton channel of the transmembrane proton pump bacteriorhodopsin. By means of

site- specific mutagenesis, single cysteine residues were introduced at 10 positions located at the protein surface, in the protein interior, and along the proton pathway. After reaction with a methanethiosulfonate spin label , the principle values of the hyperfine tensor A and the g-tensor were determined from electron paramagnetic resonance spectra measured at 170 K. The shape of the hydrophobic barrier of the proton channel is characterized in terms of a polarity index, DeltaA, determined from the variation of the hyperfine coupling constant Azz. The maximum of the hydrophobic barrier is found to be close to the retinal chromophore in the proton uptake pathway. The effect of the asymmetric distribution of charged and polar residues in the proton release and uptake pathways is clearly reflected in the behavior of the hydrophobic barrier. The presence of azide reduces the barrier height of both the cytoplasmic and extracellular channels. This finding supports the view of azide and other weakly acidic anions as catalysts for the formation of hydrogen-bonded networks in proton pathways of proteins.

Record Date Created: 19990621

33/7/9 (Item 9 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09832423 98276761 PMID: 9614577

A chimeric fusion protein containing transforming growth factor-alpha mediates gene transfer via binding to the EGF receptor.

Fominaya J; Uherek C; Wels W

Institute for Experimental Cancer Research, Tumor Biology Center, Freiburg, Germany.

Gene therapy (ENGLAND) Apr 1998, 5 (4) p521-30, ISSN 0969-7128 Journal Code: 9421525

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Fusion proteins engineered to incorporate distinct functions which co-operate in mediating the cell-type specific uptake and intracellular delivery of DNA present an attractive approach for the development of self-assembling vector systems for targeted gene transfer. Here we have chosen the EGF receptor overexpressed in many human tumors of epithelial origin as a target for a novel modular fusion protein. We have fused a cDNA fragment of the human EGF receptor ligand TGF-alpha to sequences encoding the translocation domain of Pseudomonas exotoxin A as an endosome escape activity, and the DNA-binding domain of the yeast GAL4 transcription factor. Upon bacterial expression, this TEG fusion protein displayed specific binding to EGF receptors. Complexes of the chimeric protein and plasmid DNA carrying a luciferase reporter gene, after condensation with poly-L- lysine resulted in an up to 150-fold increase in reporter gene expression in EGF receptor expressing cells in comparison to poly-L-lysine-DNA complexes alone. While in COS-1 cells no additional endosome escape activity was required, in A431 cells gene delivery was dependent on the simultaneous presence of the endosome destabilizing reagent chloroquine indicating that cell-type specific factors such as different intracellular routing of protein-DNA complexes greatly influence transfection efficiency.

Record Date Created: 19980622

33/7/10 (Item 10 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09552651 97477354 PMID: 9335564
Mapping of the residues involved in a proposed beta-strand located in the ferric enterobactin receptor FepA using site-directed spin-labeling.
Klug C S; Su W; Feix J B
Biophysics Research Institute, Medical College of Wisconsin, 8701 Watertown Plank Road, Milwaukee, Wisconsin 53226, USA.
Biochemistry (UNITED STATES) Oct 21 1997, 36 (42) p13027-33, ISSN 0006-2960 Journal Code: 0370623
Contract/Grant No.: GM22923; GM; NIGMS; GM51339; GM; NIGMS; RR01008; RR; NCRR
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Electron paramagnetic resonance (EPR) site-directed spin-labeling (SDSL) has been used to characterize a proposed transmembrane beta-strand of the Escherichia coli ferric enterobactin receptor, FepA. Each of nine consecutive residues was mutated to cysteine and subsequently labeled with the sulfhydryl-specific spin- label methanethiosulfonate (MTSL) and the purified protein reconstituted into liposomes. Continuous wave (CW) power saturation methods were used to determine exposure of the nitroxide side chains to a series of paramagnetic relaxation agents, including nickel acetylacetonate (NiAA), nickel ethylenediaminediacetate (NiEDDA), chromium oxalate (CROX), and molecular oxygen. The spin-label attached to Q245C, L247C, L249C, A251C, and Y253C had higher collision frequencies with molecular oxygen than with polar relaxation agents, indicating that these sites are exposed to the hydrophobic phase of the lipid bilayer. MTSL bound to residues S246C, E248C, E250C, and G252C had higher collision rates with the polar agents than with oxygen, suggesting that these sites are exposed to the aqueous channel. The alternating periodicity observed with the polar relaxation agents, NiAA and NiEDDA, and in opposite phase with oxygen, is consistent with beta-sheet structure. Depth measurements, based on the reciprocal concentration gradients of NiEDDA and O2 across the bilayer and calibrated for our system with phosphatidylcholine spin-labels, indicated that L249C was nearest the center of the bilayer and that Q245C and Y253C were located just below the bilayer surface in opposite leaflets of the membrane. Thus, we conclude that this approach, through mapping of individual residues, has the capability of defining beta-sheet secondary structure.
Record Date Created: 19971120

33/7/11 (Item 11 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09322895 97215828 PMID: 9062118
Kinetics and motional dynamics of spin-labeled yeast iso-1-cytochrome c: 1. Stopped-flow electron paramagnetic resonance as a probe for protein folding/unfolding of the C-terminal helix spin-labeled at cysteine 102.
Qu K; Vaughn J L; Sienkiewicz A; Scholes C P; Fetrow J S
Department of Chemistry, State University of New York at Albany, 12222, USA.
Biochemistry (UNITED STATES) Mar 11 1997, 36 (10) p2884-97, ISSN 0006-2960 Journal Code: 0370623

Contract/Grant No.: GM-35103; GM; NIGMS; GM-44829; GM; NIGMS
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The kinetics of chemically induced folding and unfolding processes in spin-labeled yeast iso-1-cytochrome c were measured by stopped-flow electron paramagnetic resonance (EPR). Stopped-flow EPR, based on a new dielectric resonator structure [Sienkiewicz, A., Qu, K., & Scholes, C. P. (1994) Rev. Sci. Instrum. 65, 68-74], gives a new temporal component to probing nanosecond molecular tumbling motions that are modulated by macromolecular processes requiring time resolution of milliseconds to seconds. The stopped-flow EPR technique presented in this work is a kinetic technique that has not been previously used with such a time resolution on spin-labeled systems, and it has the potential for application to numerous spin-labeled sites in this and other proteins. The cysteine - specific spin- label , methanethiosulfonate spin- label (MTSSL), was attached to yeast iso-1-cytochrome c at the single naturally occurring cysteine102, and the emphasis for this work was on this disulfide-attached spin-labeled prototype. This probe has the advantage of reflecting the protein tertiary fold, as shown by recent, systematic site-directed spin labeling of T4 lysozyme [Mchaourab, H. S. Lietzow, M. A., Hideg, K., & Hubbell, W. L. (1996) Biochemistry 35, 7692-7704], and protein backbone dynamics, as also shown by model peptide studies [Todd, A. P., & Millhauser, G. L. (1991) Biochemistry 30, 5515-5523]. The C-terminal cytochrome c helix where the label is attached is thought to be critical in the initial steps of protein folding and unfolding. Stopped-flow EPR resolved the monoexponential, guanidinium-induced unfolding process at pH 6.5 with an approximately 20 ms time constant; this experiment required less than 150 microL of 80 microM spin-labeled protein. We observed an approximately 50-fold decrease of this unfolding time from the 1 s range to the 20 ms time range as the guanidinium denaturant concentration was increased from 0.6 to 2.0 M. The more complex refolding kinetics of our labeled cytochrome were studied by stopped-flow EPR at pH 5.0 and 6.5. The spin probe showed a fast kinetic process compatible with the time range over which hydrogen/deuterium amide protection indicates helix formation; this process was monoexponential at pH 5.0. At pH 6.5, there was evidence of an additional slower kinetic phase resolved by stopped-flow EPR and by heme-ligation-sensitive UV-Vis that indicated a slower folding where heme misligation may be involved. Since the disulfide-attached probe has reported folding and backbone dynamics in other systems, the implication is that our kinetic experiments were directly sensing events of the C-terminal helix formation and possibly the N- and C-terminal helical interaction. The cysteine-labeled protein was also studied under equilibrium conditions to characterize probe mobility and the effect of the probe on protein thermodynamics. The difference in spin probe mobility between folded and denatured protein was marked, and in the folded protein, the motion of the probe was anisotropically restricted. The motion of the attached nitroxide in the folded protein appears to be restricted about the carbon and sulfur bonds which tether it to the cysteine. The original point of cysteine sulfur attachment is approximately 11 A from the heme iron within the C-terminal helix near its interface with the N-terminal helix, but the low-temperature EPR spin probe line width showed that the probe lies more distant (> 15 A) from the heme iron. By all physical evidence, the protein labeled at cysteine102 folded, but the spin probe in this prototype system perturbed packing which lowered the thermal melting temperature, the free energy of folding, the guanidinium concentration at the midpoint of the unfolding transition, the m parameter

of the denaturant, and the helical CD signature. This study prepares the way for study of protein folding/unfolding kinetics using EPR spectroscopy of spin- labels placed at specific cysteine -mutated sites within
Record Date Created: 19970416

33/7/12 (Item 12 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

08874398 96217790 PMID: 8641964
Decreased levels of 2-amino-3-methylimidazo[4,5-f]quinoline-DNA adducts in rats treated with beta-carotene, alpha-tocopherol and freeze-dried aloe.
Uehara N; Iwahori Y; Asamoto M; Baba-Toriyama H; Iigo M; Ochiai M; Nagao M; Nakayama M; Degawa M; Matsumoto K; Hirono I; Beppu H; Fujita K; Tsuda H
Chemotherapy Division, National Cancer Center Research Institute, Tokyo, Japan.
Japanese journal of cancer research : Gann (JAPAN) Apr 1996, 87 (4) p342-8, ISSN 0910-5050 Journal Code: 8509412
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
To assess mechanisms of chemoprevention of hepatocarcinogenesis by trans-beta-carotene (beta-C), DL-alpha-tocopherol (alpha-T), and freeze-dried whole leaves of Kidachi aloe (Aloe), formation of 2-amino-3-methylimidazo[4,5-f]quinoline (IQ)-DNA adducts was measured by 32P- post - labeling analysis, and CYP1A1 and CYP1A2 protein levels were analyzed by ELISA. Group 1 rats were fed diet containing 0.02% beta-C, 1.5% alpha-T or 30% Aloe over an 8-day period, while group 2 was given basal diet alone. On day 7, all animals were subjected to two-thirds partial hepatectomy (PH). Twelve hours after PH, they received a single dose of the carcinogenic food pyrolysate IQ (100 mg/kg) intragastrically, to initiate hepatocarcinogenesis. Rats were killed 6, 12, 24 and 48 h after IQ administration. The levels of adducts, expressed as relative adduct labeling values in rats treated with beta-C, alpha-T and Aloe, were decreased as compared with the control group at hour 24 (36 h after PH), with a significant difference in the case of the beta-C group (46.4% of the control value). Similarly, all showed a tendency for decrease at hour 48. Furthermore, the levels of CYP1A2, known to be responsible for activation of IQ, showed a significant reduction at hour 24. It is concluded that beta-C, and possibly also alpha-T and Aloe, have the potential to reduce IQ-DNA adduct formation, presumably as a result of decreased formation of active metabolites. The results may explain, at least in part, the previously observed inhibitory effects of these compounds on induction of preneoplastic hepatocellular lesions.
Record Date Created: 19960717

33/7/13 (Item 13 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

08653589 96014763 PMID: 7549753
Micronuclei and carcinogen DNA adducts as intermediate end points in nutrient intervention trial of precancerous lesions in the oral cavity.
Prasad M P; Mukundan M A; Krishnaswamy K
National Institute of Nutrition, Indian Council of Medical Research, Hyderabad, India.

European journal of cancer. Part B, Oral oncology (ENGLAND) May 1995, 31B (3) p155-9, ISSN 0964-1955 Journal Code: 9214373
Document type: Clinical Trial; Controlled Clinical Trial; Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

In cancer chemoprevention trials, biomarkers as intermediate end points have gained importance. A variety of biomarkers have been proposed as intermediate end points for upper aerodigestive tract cancers. This study was aimed at studying the frequency of micronucleated cells and carcinogen DNA adducts as indicators of DNA damage and intervention end points in chemoprevention trials. Reverse smokers of chutta (rolled tobacco) from four villages numbering 298 in total were selected. Out of these, 150 were supplemented with four nutrients (vitamin A, riboflavin, zinc and selenium) and 148 controls received placebo, one capsule twice a week for 1 year. Slides of buccal smears were prepared and stained with Fuelgen reaction and counterstained with Fast Green and examined microscopically for the presence of micronucleated cells. Oral cell washings were collected and centrifuged. The DNA adducts were evaluated by the 32P post - labelling assay method. Protein and RNA free DNA (adducted) isolated from the cells was digested with MN/SPD and the DNA adducts isolated by the butanol enrichment procedure. The DNA adducts were identified and quantitated by multidimensional chromatography on PEI-TLC sheets by screen enhanced autoradiography and presented as RAL (relative adduct labelling) values. Both the micronuclei and DNA adducts were significantly elevated in subjects with lesions. At the end of 1 year the frequency of micronuclei decreased significantly ($P < 0.001$) in the supplemented subjects with or without lesions. The DNA adducts in the supplement group at the end of 1 year also reduced significantly. The adducts decreased by 95% in subjects with all categories of lesions and by 72% in subjects without lesions. No such effects were noted in the placebo group. The two biomarkers investigated in the case study appear to be modifiable by the administration of micronutrient supplements.(ABSTRACT TRUNCATED AT 250 WORDS)

Record Date Created: 19951107

33/7/14 (Item 14 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

08455561 95210692 PMID: 7696664

Phenylglyoxal suppresses cationic lysine/K+ symport under alkaline conditions in brush border membrane vesicles from larval Manduca sexta midgut.

Parthasarathy R; Harvey W R
Department of Biology, Temple University, Philadelphia, Pennsylvania 19122.

Archives of insect biochemistry and physiology (UNITED STATES) 1995, 28 (3) p237-45, ISSN 0739-4462 Journal Code: 8501752
Contract/Grant No.: AI 30464; AI; NIAID
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The arginine-specific reagent, phenylglyoxal, decreases the initial rate of lysine/K+ symport (cotransport) as well as maximum lysine accumulation at pH 9.2, by brush border membrane vesicles obtained from the larval

midgut of the lepidopteran, Manduca sexta. The symport of a neutral amino acid, leucine, remained unaffected. Following exposure to phenylglyoxal, the apparent dissociation constant for lysine increased by a factor of 2.5 whereas the maximum uptake rate decreased by a factor of 0.4. More than one arginine residue appears to react with phenylglyoxal. Apparently phenylglyoxal reacts preferentially with arginine residues on a symporter that is specific for positively charged lysine . Phenylglyoxal shows promise as a specific covalent label for the identification of a cationic amino acid symporter.
Record Date Created: 19950502

33/7/15 (Item 15 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

07897154 94032335 PMID: 8218279
Formation of the meta II photointermediate is accompanied by conformational changes in the cytoplasmic surface of rhodopsin.
Resek J F; Farahbakhsh Z T; Hubbell W L; Khorana H G
Department of Biology, Massachusetts Institute of Technology, Cambridge 02139.
Biochemistry (UNITED STATES) Nov 16 1993, 32 (45) p12025-32, ISSN 0006-2960 . Journal Code: 0370623
Contract/Grant No.: AI 11479; AI; NIAID; EYO6189; EY; NEI; GM 28289; GM; NIGMS; +
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Five mutations of rhodopsin have been produced, each of which contains a unique cysteine residue at positions 62, 65, 140, 240, or 316 in the cytoplasmic domain. The single reactive cysteines were derivatized with a sulfhydryl- specific nitroxide spin- label , and the electron paramagnetic resonance (EPR) spectra were analyzed in both lauryl maltoside and digitonin in the dark and after photobleaching. The collision rate of the attached nitroxides with polar and nonpolar paramagnetic agents indicated that they were all exposed to the aqueous environment. Photobleaching of the mutants in digitonin, which arrests the protein at the meta I intermediate, produced little change in mobility of the attached nitroxide. On the other hand, photobleaching in lauryl maltoside produced the meta II intermediate and significant changes in the EPR spectra of the nitroxides attached to positions 140 and 316. These data directly reveal a light-induced conformational change in the cytoplasmic loops that accompanies meta II formation.
Record Date Created: 19931221

33/7/16 (Item 16 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

06579723 90276439 PMID: 2351140
Calcium-dependent distance changes in binary and ternary complexes of troponin.
Schulzki H D; Kramer B; Fleischhauer J; Mercola D A; Wollmer A
Lehr- und Forschungsgebiet Struktur und Funktion der Proteine, Institut fur Biochemie, Rheinisch-Westfalische Technische Hochschule Aachen, Federal Republic of Germany.

European journal of biochemistry / FEBS (GERMANY, WEST) May 20 1990, 189 (3) p683-92, ISSN 0014-2956 Journal Code: 0107600
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

Calcium-dependent distance changes have been determined by resonance energy transfer in binary and ternary troponin complexes in order to collect evidence for the structural rearrangements which are part of the hypothetical trigger mechanism of skeletal muscle contraction. Donor and acceptor fluorophores were either intrinsic tryptophans in subunits with a favourable sequence from different species, quasi-intrinsic Tb3+ ions bound to troponin C or extrinsic labels attached to specific cysteine or methionine residues. All chemically modified subunits proved fully active in conferring calcium sensitivity onto myosin ATPase. Nine distances were determined between five sites which allowed construction of a three-dimensional lattice representing the spatial distribution of four sites in the ternary complex of troponin C, I and T. Distances in binary complexes were nearly unaltered upon addition of the third subunit. Regulatory calcium binding caused distance changes of the order of 0.7-1.1 nm. In view of the large displacements of the hypothetical mechanism, they turned out to be smaller than anticipated. The fluorophoric sites selected may be localized in a zone of the troponin complex which happens to be relatively little affected by the mechanism. Alternatively, amplification of the moderate changes seen here would require the complete set of thin filament proteins.

Record Date Created: 19900713

33/7/17 (Item 17 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

06402368 90090059 PMID: 2480832

Slow component B protein kinetics in optic nerve and tract windows.

Paggi P; Lasek R J; Katz M J
Bio-architecronics Center, School of Medicine, Case Western Reserve University, Cleveland, OH 44106.

Brain research (NETHERLANDS) Dec 18 1989, 504 (2) p223-30, ISSN 0006-8993 Journal Code: 0045503
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The transport kinetics of 3 radiolabeled slow component b (SCb) proteins (a 30 kDa protein, clathrin, and actin) were examined in the axons of mouse retinal ganglion cells. To view the transit of these proteins through the entire optic pathway between the eye and the target cells, we used two different windows: (1) a 2 mm segment from the optic nerve located 3-5 mm from the eye, and (2) a 2 mm segment from the optic tract located past the chiasm 6-8 mm from the eye. The radiolabeled proteins from these windows were separated by 1- and 2-dimensional SDS-PAGE, and the individual radiolabeled bands were quantified. Radiolabeled proteins entered and cleared the optic axons between 1 and 119 days post - labeling . All these proteins had broader transport waves in the more distal optic tract window than in the more proximal optic nerve window. The spreading of transport waves as they advance along the axon appears to be produced by a playing out of the natural heterogeneity of axonal transport rates within

each population of labeled proteins. Our results confirm the proposals that clathrin and the 30 kDa protein are transported principally with SCb and that actin is transported both with SCb and with SCa. Although these proteins can be generally classified with SCb, their detailed kinetics differed (for example, their median transit times differed) and, in summary, their characteristic rates of movement can be ordered as: clathrin greater than 30 kDa protein greater than actin.(ABSTRACT TRUNCATED AT 250 WORDS)
Record Date Created: 19900206

33/7/18 (Item 18 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

05727520 88151973 PMID: 3345755
The reconstitution of a hybrid histone octamer containing avian 110Cys-des-thio-histone H3 and sea-urchin 73Cys-histone H4.
Greyling H J; Sewell B T; Von Holt C
Department of Biochemistry, University of Cape Town, Rondebosch, South Africa.
European journal of biochemistry / FEBS (GERMANY, WEST) Feb 1 1988, 171 (3) p721-6, ISSN 0014-2956 Journal Code: 0107600
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A hybrid histone octamer was reconstituted from erythrocyte H2A and H2B, avian [110 Cys-des-thio]histone H3 and the sea-urchin sperm [73Cys]H4 variant. [110Cys-Des-thio]histone H3 was prepared by reaction of natural H3 with Raney nickel. The ability of the hybrid octamer to crystallize to the same form as the natural octamer demonstrated that the chemical modification of cysteine to alanine in H3 and the mutation from threonine to cysteine in sperm H4 do not alter histone-histone interactions in the octamer. Since the sulfhydryl groups of both H4 molecules are fully accessible to 5,5'-dithiobis(2-nitrobenzoate) these residues provide suitable sites for the introduction of a single cysteine - specific label per H4 molecule in the octamer.
Record Date Created: 19880411

33/7/19 (Item 19 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

05710341 88139240 PMID: 2449424
The recovery of slow axonal transport after a single intraperitoneal injection of beta, beta'-iminodipropionitrile in the rat.
Komiya Y; Cooper N A; Kidman A D
Neurobiology Unit, School of Life Sciences, New South Wales Institute of Technology, Gore Hill, Australia.
Journal of biochemistry (JAPAN) Oct 1987, 102 (4) p869-73, ISSN 0021-924X Journal Code: 0376600
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A single intraperitoneal injection of beta, beta'-iminodipropionitrile (IDPN) at a dose of 1.5 g/kg was given to 4-week-old rats. Immediately

following, or at 1, 2, 3, 5, 10, and 15 weeks after IDPN injection, [35S]methionine was introduced into the anterior horn area of the lumbar cord. Labeled axonal proteins in the sciatic nerve were analyzed electrophoretically and fluorographically at 5, 10, and 15 weeks post - labeling . Labeled neurofilament proteins halt for a short period just after IDPN injection, then continue migrating distally, though at a slower rate, and finally the transport of affected neurofilament proteins completely recovers by 6 weeks post - labeling .
Record Date Created: 19880325

33/7/20 (Item 20 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

05603432 88029413 PMID: 3311742
Fluorescent labeling of cysteinyl residues. Application to extensive primary structure analysis of proteins on a microscale.
Gorman J J; Corino G L; Mitchell S J
Commonwealth Scientific and Industrial Research Organisation, Australian Animal Health Laboratory, Geelong.
European journal of biochemistry / FEBS (GERMANY, WEST) Oct 1 1987, 168 (1) p169-79, ISSN 0014-2956 Journal Code: 0107600
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The specificity and efficiency of fluorescent labeling of proteins by reduction and subsequent alkylation with 5-N-[(iodoacetamidoethyl)amino]naphthalene-1-sulfonic acid (5-I-AEDANS) [J.J. Gorman, (1987) Anal. Biochem. 160, 376-387] has been investigated. Proteins studied include porcine insulin, chicken ovalbumin and bovine serum albumin. Amino acid analysis of the B-chain derivative of insulin revealed quantitative recovery of cysteine in its S-carboxymethyl form and no other carboxymethylated amino acid derivatives. Fast-atom-bombardment mass spectrometric (FAB-MS) analysis of this derivative also indicated specific labeling of cysteine residues and automated stepwise protein sequence analysis of the derivative was performed to completion with initial and average repetitive yields of 73% and 96%, respectively. Tryptic peptides produced from the ovalbumin and serum albumin derivatives were fractionated by HPLC and subsequently analysed by amino acid analysis, FAB-MS and automated stepwise protein sequence analysis. These analyses have revealed that the labeling procedure exhibits a high degree of efficiency and is specifically directed towards S-alkylation of cysteine residues. The high level of fluorescence intensity of the label enabled specific detection of trace quantities of cysteine -containing peptides derived from contaminating protein(s). It is apparent that in addition to facilitating isolation of small quantities of proteins the labeling procedure is compatible with standard protein chemistry techniques involved in obtaining extensive structural data on isolated proteins.
Record Date Created: 19871207

33/7/21 (Item 21 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

05202990 86274472 PMID: 2874017
Effects of cysteamine administration on the in vivo incorporation of

[35S] cysteine into somatostatin-14, somatostatin-28, arginine vasopressin, and oxytocin in rat hypothalamus.
Cameron J L; Fernstrom J D
Endocrinology (UNITED STATES) Sep 1986, 119 (3) p1292-7, ISSN 0013-7227 Journal Code: 0375040
Contract/Grant No.: HD-20887; HD; NICHD; MH-00254; MH; NIMH; NS20017; NS; NINDS
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The effect of cysteamine injection on the in vivo incorporation of [35S] cysteine into somatostatin-14 (SRIF-14), SRIF-28, arginine vasopressin (AVP), and oxytocin (OXT) in rat hypothalamus was studied. [35S] Cysteine was injected into the third ventricle 1 h, 4 h, or 1 week after cysteamine (300 mg/kg, sc) injection; animals were killed 4 h later. The drug was found to substantially reduce immunoreactive SRIF levels, but not OXT or AVP, 4 h after its injection. Cysteamine also caused large reductions in label incorporation into SRIF-14, SRIF-28, and OXT 1 and 4 h after drug injection. However, [35S] cysteine incorporation into AVP was increased substantially at these time points, while that into acid-precipitable protein was normal. One week after cysteamine injection, label incorporation into all hypothalamic peptides was normal. Cysteine specific activity was also measured after [35S] cysteine injection and was found to be similar in treatment and control groups. The results suggest that cysteamine inhibits the syntheses of SRIF-14, SRIF-28, and OXT and stimulates that of AVP.
Record Date Created: 19860919

33/7/22 (Item 22 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

05034349 86088958 PMID: 3941405
Carboxyl-modified amino acids and peptides as protease inhibitors.
Thompson S A; Andrews P R; Hanzlik R P
Journal of medicinal chemistry (UNITED STATES) Jan 1986, 29 (1) p104-11, ISSN 0022-2623 Journal Code: 9716531
Contract/Grant No.: GM-07775; GM; NIGMS
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Several types of carboxyl-modified amino acids and peptides were prepared in forms having N-terminal modifications (carrier fragments) suitable for one of several representative protease enzymes, and their inhibitory action toward those enzymes were evaluated. The carboxyl modifications (inhibitory units) included (b) CONH2, (c) CSNH2, (d) CN, (e) trans-CH = CHCO2Me, and (f) trans-CH = CHSO2Me. The carrier fragments included NH2(PhCH2)CHX (1), AcNH(PhCH2)CHX (2), H2NCH2CONH(PhCH2)CHX (3), and AcNH(PhCH2)CHCONHCH2X (4). Compounds 1b, 1d, 1e, and 1f were competitive inhibitors of both microsomal and cytosolic leucine aminopeptidase (Ki = 14.8, 67, 61, and 3.7 mM with the former and 14.1, 26.4, 27.3, and 8.8 mM with the latter, respectively). Neither compound 1c nor leucine thioamide had any detectable effect on either enzyme. Compounds 2b-f were also competitive inhibitors toward chymotrypsin (Ki = 13.9, 23.0, 5.3, 30.8, and 29.4 mM, respectively). While 4b, 4c, and 4d were competitive inhibitors of papain

(Ki = 4.7, 0.095, and 0.0011 mM, respectively), 4e proved to be an irreversible affinity label (Ki = 0.026 mM and k2 = 0.0018 s-1). Inactivation of papain by 4e was retarded in the presence of 4d and could not be reversed by dialysis. Similarly 3b and 3d were competitive inhibitors of dipeptidyl aminopeptidase I (DPP-I, EC 3.4.14.1) (Ki = 6.2 and 0.0027 mM, respectively), while 3e and 3f were irreversible affinity labels (Ki = 0.22 and 0.18 mM, and k2 = 0.015 and 0.010 s-1, respectively). Inhibition of DPP-I by 3d provides only the second example of a cysteine protease which is strongly inhibited by a nitrile analogue of a specific substrate. Further studies are needed to determine the generality and potential utility of this finding. Compounds 3e, 3f, and 4e exemplify a new class of specific affinity labels for cysteine proteases whose activity probably derives from irreversible Michael addition of the catalytic cysteine to the activated double bond.

Record Date Created: 19860219

33/7/23 (Item 23 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

04988232 86059349 PMID: 2415512

Labeling of specific lysine residues at the active site of glutamine synthetase.

Colanduoni J; Villafranca J J

Journal of biological chemistry (UNITED STATES) Dec 5 1985, 260 (28) p15042-50, ISSN 0021-9258 Journal Code: 2985121R

Contract/Grant No.: GM23529; GM; NIGMS; RR01412; RR; NCRR

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Glutamine synthetase (Escherichia coli) was incubated with three different reagents that react with lysine residues, viz. pyridoxal phosphate, 5'-p-fluorosulfonylbenzoyladenosine, and thiourea dioxide. The latter reagent reacts with the epsilon-nitrogen of lysine to produce homoarginine as shown by amino acid analysis, nmr, and mass spectral analysis of the products. A variety of differential labeling experiments were conducted with the above three reagents to label specific lysine residues. Thus pyridoxal phosphate was found to modify 2 lysine residues leading to an alteration of catalytic activity. At least 1 lysine residue has been reported previously to be modified by pyridoxal phosphate at the active site of glutamine synthetase (Whitley, E. J., and Ginsburg, A. (1978) J. Biol. Chem. 253, 7017-7025). By varying the pH and buffer, one or both residues could be modified. One of these lysine residues was associated with approximately 81% loss in activity after modification while modification of the second lysine residue led to complete inactivation of the enzyme. This second lysine was found to be the residue which reacted specifically with the ATP affinity label 5'-p-fluorosulfonylbenzoyladenosin e. Lys-47 has been previously identified as the residue that reacts with this reagent (Pinkofsky, H. B., Ginsburg, A., Reardon, I., Heinrikson, R. L. (1984) J. Biol. Chem. 259, 9616-9622; Foster, W. B., Griffith, M. J., and Kingdon, H. S. (1981) J. Biol. Chem. 256, 882-886). Thiourea dioxide inactivated glutamine synthetase with total loss of activity and concomitant modification of a single lysine residue. The modified amino acid was identified as homoarginine by amino acid analysis. The lysine residue modified by thiourea dioxide was established by differential labeling experiments to be the same residue associated with the 81% partial

loss of activity upon pyridoxal phosphate inactivation. Inactivation with either thiourea dioxide or pyridoxal phosphate did not affect ATP binding but glutamate binding was weakened. The glutamate site was implicated as the site of thiourea dioxide modification based on protection against inactivation by saturating levels of glutamate. Glutamate also protected against pyridoxal phosphate labeling of the lysine consistent with this residue being the common site of reaction with thiourea dioxide and pyridoxal phosphate.
Record Date Created: 19860103

33/7/24 (Item 24 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

04872861 85252857 PMID: 3160389
Photoaffinity labeling of functionally different lysine-binding sites in human plasminogen and plasmin.
Ryan T J; Keegan M C
Biochimica et biophysica acta (NETHERLANDS) Aug 8 1985, 830 (2) p187-94, ISSN 0006-3002 Journal Code: 0217513
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Photoaffinity labeling of human plasmin using 4-azidobenzoylglycyl-L-lysine inhibits clot lysis activity, while the activity toward the active-site titrant, p-nitrophenyl-p'-guanidinobenzoate, or alpha-casein are maintained. Photoaffinity labeling of native Glu-plasminogen with the same reagent causes incorporation of approximately 1.5 mol label per mol plasminogen. This labeled plasminogen can be activated to plasmin by either urokinase or streptokinase. The resulting plasmin has full clot lysis activity and can be subsequently photoaffinity labeled with a loss of clot lysis activity. The rate of activation of labeled plasminogen by urokinase is increased relative to that of native plasminogen. epsilon-Aminocaproic acid blocks incorporation of photoaffinity label into both plasminogen and plasmin, indicating that the labeling is specific to the lysine -binding sites. The labels are located in the kringle 1+2+3 fragment in either photoaffinity-labeled plasminogen or plasmin. These results indicate that the specific lysine-binding site blocked in plasmin acts in concert with the active-site in binding and using fibrin as a substrate. This clot lysis regulating site is not available for labeling in plasminogen, but is exposed or changed upon activation to plasmin. The different lysine-binding sites labeled in plasminogen may regulate the conformation of the molecule as evidence by an enhanced rate of activation to plasmin.
Record Date Created: 19850912

33/7/25 (Item 25 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

04478552 84160755 PMID: 6706430
Factors affecting the labeling of human fibrinogen with 99mTc in vitro.
Lavie E; Shenberg C; Bitton M; Mechlis S
International journal of applied radiation and isotopes (UNITED STATES) Feb 1984, 35 (2) p99-102, ISSN 0020-708X Journal Code: 0374715
Document type: Journal Article
Languages: ENGLISH

Main Citation Owner: NLM
Record type: Completed
The preparation of [99mTc]fibrinogen is described. The following factors which could affect the quality of this preparation were studied: pH, SnCl2 and protein concentrations, pre- and post - labeling incubation times and labeling temperature. A 90% labeling yield was achieved by incubating fibrinogen with SnCl2 at pH 9.8 for 22 h before adding the TcO-4 solution. The product obtained was stable and was 85% clottable. The x-ray fluorescence technique was employed to measure the amount of bound tin at different times.
Record Date Created: 19840517

33/7/26 (Item 26 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

04146317 83153076 PMID: 6131670
The rate of protein degradation in developing brain. Methodological considerations.
Dunlop D S; McHale D M; Lajtha A
Biochemical journal (ENGLAND) Dec 15 1982, 208 (3) p659-66, ISSN 0264-6021 Journal Code: 2984726R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Recently we reported that the rate of protein breakdown decreases during development. Breakdown rates were calculated from the rates of protein synthesis and the changes in brain protein content with age. A different study, measuring breakdown by monitoring the loss of label from brain protein after an H14CO3- pulse, came to the opposite conclusion: that the rate of breakdown is low in immature brain and increases during development. We have now investigated some of the factors (the distribution of label in protein and the potential for recycling) that might introduce errors into these measurements . The specific radioactivities of both protein-bound and free amino acids were determined in the brains of young rats several days after an intraperitoneal pulse of H14CO3-. For a number of amino acids the specific radioactivity of the free amino acid is high compared with that of the protein-bound amino acid, and therefore recycling could result in an underestimate of the degradation rate. Because glutamic acid had a relatively low specific -radioactivity ratio, [1-14C] glutamic acid was used in a pulse-labelling experiment to measure degradation. The rate so obtained, 0.6% . h-1, is twice the rate found with H14CO3- labelling (based on total protein-bound radioactivity). Insofar as recycling is a possible complication, 0.6% . h-1 may be a minimum value. Although somewhat higher degradation rates are found after labelling with an intracranial pulse, which was considered as a possible route to limit recycling, there are difficulties in interpreting these data.
Record Date Created: 19830421

33/7/27 (Item 27 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

04037239 83030766 PMID: 7130164
Active site studies of cytochrome P=450CAM. I. Specific cysteine labeling with the affinity reagent isobornyl bromoacetate as a model for substrate

binding.
Murray R I; Gunsalus I C; Dus K M
Journal of biological chemistry (UNITED STATES) Nov 10 1982, 257 (21) p12517-25, ISSN 0021-9258 Journal Code: 2985121R
Contract/Grant No.: AM-00562; AM; NIADDK; GM-21161; GM; NIGMS; GM-21726; GM; NIGMS
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A model is presented suggesting a function of specific cysteine residue of cytochrome P-450CAM in binding the substrate camphor, via a thiohemiketal bond, for its correct orientation to the heme iron and for the subsequent transfer of nascent product to facilitate its release. This model was developed to explain the results of affinity labeling with isobornyl bromoacetate. This reagent couples to the proteins via a thioether bond to cysteine, eliciting a type I transition in the difference spectrum. Formation of this covalent complex, which is strongly inhibited by the substrate, can be monitored by quantitation of S-carboxymethylcysteine in acid hydrolyzates. While addition of one equivalent of label yields 0.3 equivalents of the cysteine derivative after 5 min, increasing to 0.8 equivalents after 24 h, the spectral shift decays with time. Kinetic analysis of the spectral decay and of covalent coupling strongly suggests that thioether bond formation occurs at the substrate binding-site, in a reaction step prior to, and distinct from, the step associated with the spectral decay. The P-450CAM derivative, when titrated with camphor, produced again a type I spectrum virtually identical with the spectrum of the native P-450CAM-substrate complex. While the model presented here is not the only possible interpretation of the results, it is fully consistent with them and provides an excellent framework for further study of the catalytic mechanism of P-450CAM.
Record Date Created: 19821218

33/7/28 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13270409 BIOSIS NO.: 200100477558
Electron spin resonance and fluorescence studies of the bound-state conformation of a model protein substrate to the chaperone SecB.
AUTHOR: Panse Vikram G; Beena K; Philipp Reinhard; Trommer Wolfgang E; Vogel Pia D; Varadarajan Raghavan(a)
AUTHOR ADDRESS: (a)Molecular Biophysics Unit, Indian Institute of Science, Bangalore, 560 012: varadar@mbu.iisc.ernet.in**India
JOURNAL: Journal of Biological Chemistry 276 (36):p33681-33688 September 7, 2001
MEDIUM: print
ISSN: 0021-9258
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: SecB is a homotetrameric, cytosolic chaperone that forms part of the protein translocation machinery in Escherichia coli. We have investigated the bound-state conformation of a model protein substrate of

SecB, bovine pancreatic trypsin inhibitor (BPTI) as well as the conformation of SecB itself by using proximity relationships based on site-directed spin-labeling and pyrene fluorescence methods. BPTI is a 58-residue protein and contains three disulfide groups between residues 5 and 55, 14 and 38, as well as 30 and 51. Mutants of BPTI that contained only a single disulfide were reduced, and the free cysteines were labeled with either thiol- specific spin labels or pyrene maleimide. The relative proximity of the labeled residues was studied using either electron spin resonance spectroscopy or fluorescence spectroscopy. The data suggest that SecB binds a collapsed coil of reduced unfolded BPTI, which then undergoes a structural rearrangement to a more extended state upon binding to SecB. Binding occurs at multiple sites on the substrate, and the binding site on each SecB monomer accommodates less than 21 substrate residues. In addition, we have labeled four solvent-accessible cysteine residues in the SecB tetramer and have investigated their relative spatial arrangement in the presence and absence of the substrate protein. The electron spin resonance data suggest that these cysteine residues are in close proximity (15 ANG) when no substrate protein is bound but move away to a distance of greater than 20 ANG when SecB binds substrate. This is the first direct evidence of a conformational change in SecB upon binding of a substrate protein.

33/7/29 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13157189 BIOSIS NO.: 200100364338
Detection of fluorescence dye-labeled proteins in 2-D gels using an ArthurTM 1442 Multiwavelength Fluoroimager.
AUTHOR: Herick Klaus(a); Jackson Peter; Wersch Gregor; Burkovski Andreas
AUTHOR ADDRESS: (a)PerkinElmer Life Sciences, Rolandplatz 1a, D-76135, Karlsruhe: klaus.herick@perkinelmer.com**Germany
JOURNAL: Biotechniques 31 (1):p146-149 July, 2001
MEDIUM: print
ISSN: 0736-6205
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: Labeling of proteins with SYPRO(R) Orange, SYPRO Red, and SYPRO Ruby after 2-D polyacrylamide gel electrophoresis (PAGE) using plastic-backed immobilized pH gradient (IPG) strips and precast SDS polyacrylamide gels was tested. Protein spots were detected using an ArthurTM 1442 Multiwavelength Fluoroimager. The labeling methods described allow detection of proteins both after isoelectric focusing (IEF) and PAGE with a sensitivity higher than or comparable to standard silver staining methods. In addition to the post - labeling methods mentioned above, pre-labeling with the cysteine - specific fluorophore monobromobimane before 2-D PAGE is a sensitive, fast, and cost-effective alternative to existing staining protocols.

33/7/30 (Item 3 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13049668 BIOSIS NO.: 200100256817
Concerted movement of transmembrane helices coupled to activation of a peptide hormone GPCR.
AUTHOR: Karnik Sadashiva S(a); Miura Shin-ichiro(a); Boros John(a)
AUTHOR ADDRESS: (a)Lerner Res. Institute, The Cleveland Clinic, 9500 Euclid Ave., Cleveland, OH, 44195**USA
JOURNAL: FASEB Journal 15 (5):pA1165 March 8, 2001
MEDIUM: print
CONFERENCE/MEETING: Annual Meeting of the Federation of American Societies for Experimental Biology on Experimental Biology 2001 Orlando, Florida, USA March 31-April 04, 2001
ISSN: 0892-6638
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: A key step in transmembrane (TM) signal transduction by G protein coupled receptors (GPCR) is the ligand-induced conformational change of the receptor which triggers the activation of a specific cytoplasmic guanine nucleotide-binding protein. GPCRs contain a seven TM-helical structure essential for signal transduction in response to a large variety of sensory and hormonal signals. Studies of rhodopsin, biogenic amine and peptide GPCRs have shown that the second TM segment contains a highly conserved Asp residue which is critical for agonist-activation in these receptors. However, the role of conformational changes in TM2 to signal transduction by a GPCR is not known because activation-induced conformational changes in TM2 have not been measured . Here we use modification of reporter cysteines to measure water accessibility at specific residues in TM2 and TM7 of the type 1 receptor for the octapeptide hormone angiotensin II. Activation-dependent accessibility changes were measured at several consecutive residues on TM2 and TM7 in a constitutively activated mutant. The accessibility changes directly correlated with degree of activation, establishing the link between physical changes in TM-helices and function. Measured accessibility changes suggest that the TM2 and TM7 undergo a transmembrane movement in response to activation. This is the first report of in situ measurement of TM2 movement in a GPCR. A model for movement of helices responsible for receptor activation will be discussed.

33/7/31 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

11294127 BIOSIS NO.: 199800075459
Interaction of cytochrome c and cytochrome c oxidase studied by spin-label EPR and site-directed mutagenesis.
AUTHOR: Park Hee Young; Chun Sun Bum; Han Sanghwa(a); Lee Kwangsoon; Kim Kyunghoon
AUTHOR ADDRESS: (a)Dep. Biochemistry, Coll. Natural Sci., Kangwon Natl. Univ., Chunchon 200-701**South Korea
JOURNAL: Journal of Biochemistry and Molecular Biology 30 (6):p397-402 Nov. 30, 1997
ISSN: 1225-8687
DOCUMENT TYPE: Article
RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: A thiol- specific spin label was attached to cysteine -102 of yeast cytochrome c and electron paramagnetic resonance (EPR) spectra were measured as a function of added cytochrome c oxidase concentration. The intensity decreased due to line broadening as cytochrome c formed a complex with cytochrome c oxidase and reached a minimum when the ratio of cytochrome c to cytochrome c oxidase became one. Replacement of either Lys-72 or Lys-87 of cytochrome c by Glu did not result in a significant change in binding affinity. Interestingly the K72E mutant, unlike K87E, had a much lower rate of electron transfer than the wild type. These results indicate that many positively charged residues as a group participate in complex formation but Lys-72 might be important for cytochrome c to be locked in an orientation for an efficient electron transfer. A stoichiometry of 1 was also confirmed by optical absorption of the cytochrome c-cytochrome c oxidase complex which had been run through a gel chromatography column to remove unbound cytochrome c. The EPR spectrum of this 1:1 complex, however, was a mixture of two components. This explains a biphasic kinetics for a single binding site on cytochrome c oxidase without invoking conformational transition.

33/7/32 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

10528979 BIOSIS NO.: 199699150124
EPR spectra of spin-labeled cytochrome c bound to acidic membranes: Implications for the binding site and reversibility.
AUTHOR: Min Tongpil; Park Nan-Hyang; Park Hee Young; Hong Sun-Joo; Han Sanghwa
AUTHOR ADDRESS: Dep. Biochemistry, Kangwon Natl. Univ., Chunchon 200-701** South Korea
JOURNAL: Journal of Biochemistry and Molecular Biology 29 (2):p169-174 1996
ISSN: 1225-8687
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Yeast cytochrome c (cyt c) was modified at cysteine -102 with a thiol- specific spin label and its interaction with liposomes containing acidic phospholipids was studied by electron paramagnetic resonance (EPR) spectroscopy. Association of cyt c with liposomes resulted in a significant reduction in the mobility of the spin label and a fraction of cyt c even seemed to be immobilized. Based on a large spectral change upon binding and the proximity of the spin-label to lysine-86 and -87, we propose these two residues to be the potential binding site at neutral pH. The interaction is electrostatic in nature because the spectral changes were reversed by addition of anions. Dissociation of the bound cyt c by anions, however, became less effective as the lipid/protein ratio increased. This suggests a repulsive lateral interaction among the bound cyt c. Unlabeled cyt c molecules added to preformed cyt c-liposome complex displaced the bound (spin labeled) cyt c and the process was competitive and reversible.

33/7/33 (Item 6 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

10146000 BIOSIS NO.: 199698600918
Synthesis of two series of cysteine - specific spin labels containing a 15N substituted nitroxide.
AUTHOR: Wu Ge
AUTHOR ADDRESS: Dep. Anesthesia, Mass. General Hosp., Boston, MA 02114** USA
JOURNAL: Journal of Labelled Compounds and Radiopharmaceuticals 36 (12):p 1173-1181 1995
ISSN: 0362-4803
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Synthesis of two series of spin labels with a cysteine-reactive group, either maleimide or methanethiosulfonate, and a 15N substituted nitroxide separated at variable distance is reported. In the synthesis, the cysteine-reactive groups were first incorporated into the target molecule and the 15N substituted nitroxide was introduced in the last step via a mild reaction without destroying the cysteine-reactive groups. The synthetic routes described here are applicable to the synthesis of spin labels with either 15N substituted or "normal" 14N nitroxide. Spin labeling of cysteine residues in rhodopsin by the newly synthesized spin labels was also demonstrated.

33/7/34 (Item 7 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

07261505 BIOSIS NO.: 000090041381
CALCIUM-DEPENDENT DISTANCE CHANGES IN BINARY AND TENARY COMPLEXES OF TROPONIN
AUTHOR: SCHULZKI H-D; KRAMER B; FLEISCHHAUER J; MERCOLA D A; WOLLMER A
AUTHOR ADDRESS: LEHR- UND FORSCHUNGSGEBIET STRUKTUR UND FUNKTION PROTEINE, INST. BIOCHEM., RHEINISCH-WESTFAELISCHE TECHNISCHE HOCHSCHULE AACHEN, KLIN. PAUWELSSTR. 30, D-5100 AACHEN, FRG.
JOURNAL: EUR J BIOCHEM 189 (3). 1990. 683-692. 1990
FULL JOURNAL NAME: European Journal of Biochemistry
CODEN: EJBCA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Calcium-dependent distance changes have been determined by resonance energy transfer in binary and ternary troponin complexes in order to collect evidence for the structural rearrangments which are part of the hypothetical trigger mechanism of skeletal muscle contraction. Donor and acceptor fluorophores were either intrinsic tryptophans in subunits with a favourable sequence from different species, quasi-intrinsic Tb3+ ions bound to troponin C or extrinic labels attached to specific cysteine or methionine residues. All chemically modified subunits proved fully active in conferring calcium sensitivity onto myosin ATPase. Nine distances were determined between five sites which allowed construction of a three-dimensional lattice representing

the spatial distribution of four sites in the ternary complex of troponin C, I and T. Distances in binary complexes were nearly unaltered upon addition of the third subunit. Regulatory calcium binding caused distance changes of the order of 0.7-1.1 nm. In view of the large displacements of the hypothetical mechanism, they turned out to be smaller than anticipated. The fluorophoric sites selected may be localized in a zone of the troponin complex which happens to be relatively little affected by the mechanism. Alternatively, amplification of the moderate changes seen here would require the complete set of thin filament proteins.

33/7/35 (Item 8 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

07240008 BIOSIS NO.: 000090019883
SYNTHESIS OF CARBON-14 AND TRITIUM LABELED METHYLPREDNISOLONE SULEPTANATE
AUTHOR: STOLLE W T; RUNGE T A; HSI R S P
AUTHOR ADDRESS: RES. LAB. THE UPJOHN COMPANY, KALAMAZOO, MI 49007, USA.
JOURNAL: J LABELLED COMPD RADIOPHARM 28 (5). 1990. 555-580. 1990
FULL JOURNAL NAME: Journal of Labelled Compounds & Radiopharmaceuticals
CODEN: JLCRD
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Methylprednisolone suleptanate was initially labeled with tritium in the A-ring of the steroid portion of the molecule, and with carbon-14 at both carboxylic carbons of the suberic acid portion of the side chain. However these labels proved to lack total metabolic stability after administration to rats. Subsequently a second pair of labeled methylprednisolone suleptanates was synthesized, with tritium at C-7 in the B-ring of the steroid and carbon-14 exclusively at the carboxamide carbon in the side chain. These labeled compounds showed excellent metabolic stability of both the tritium and carbon-14 labels, and should be well suited for conducting drug disposition studies.

33/7/36 (Item 9 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

06134268 BIOSIS NO.: 000085097420
THE RECONSTITUTION OF A HYBRID HISTONE OCTAMER CONTAINING AVIAN CYSTEINE-110-DES-THIO-HISTONE H3 AND SEA-URCHIN CYSTEINE-73-HISTONE H4
AUTHOR: GREYLING H J; SEWELL B T; VON HOLT C
AUTHOR ADDRESS: UCT-CSIR RES. CEN. MOL. BIOL., DEP. BIOCHEM., UNIV. CAPE TOWN, PRIVATE BAG, RONDEBOSCH, S. AFR. 7700.
JOURNAL: EUR J BIOCHEM 171 (3). 1988. 721-726. 1988
FULL JOURNAL NAME: European Journal of Biochemistry
CODEN: EJBCA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: A hybrid histone octamer was reconstituted from erythrocyte H2A and H2B, avian [110Cys-des-thio]histone H3 and the sea-urchin sperm [73Cys]H4 variant. [110Cys-Des-thio]histone H3 was prepared by reaction of natural H3 with Raney nicke. The ability of the hybrid octamer to

crystallise to the same form as the natural octamer demonstrated that the chemical modification of cysteine to alanine in H3 and the mutation from threonine to cysteine in sperm H4 do not alter histone-histone interactions in the octamer. Since the sulfhydryl groups of both H4 molecules are fully accessible to 5,5'-dithiobis(2-nitrobenzoate) these residues provide suitable sites for the introduction of a single cysteine - specific label per H4 molecule in the octamer.

33/7/37 (Item 10 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

05260787 BIOSIS NO.: 000082101412
EFFECTS OF CYSTEAMINE ADMINISTRATION ON THE IN-VIVO INCORPORATION OF SULFUR-35 CYSTEINE INTO SOMATOSTATIN-14 SOMATOSTATIN-28 ARGININE VASOPRESSIN AND OXYTOCIN IN RAT HYPOTHALAMUS
AUTHOR: CAMERON J L; FERNSTROM J D
AUTHOR ADDRESS: WESTERN PSYCHIATRIC INSTITUTE AND CLINIC, 3811 O'HARA STREET, PITTSBURGH, PENNSYLVANIA 15213.
JOURNAL: ENDOCRINOLOGY 119 (3). 1986. 1292-1297. 1986
FULL JOURNAL NAME: Endocrinology
CODEN: ENDOA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The effect of cysteamine injection on the in vivo incorporation of [35S] cysteine into somatostatin-14 (SRIF-14), SRIF-28, arginine vasopressin (AVP), and oxytocin (OXT) in rat hypothalamus was studied. [35S] Cysteine was injected into the third ventricle 1 h, 4 h, or 1 week after cysteamine (300 mg/kg, sc) injection; animals were killed 4 h later. The drug was found to substantially reduce immunoreactive SRIF levels, but not OXT or AVP, 4 h after its injection. Cysteamine also caused large reductions in label incorporation into SRIF-14, SRIF-28, and OXT 1 and 4 h after drug injection. However, [35S] cysteine incorporation into AVP was increased substantially at these time points, while that into acid-precipitable protein was normal. One week after cysteamine injection, label incorporation into all hypothalamic peptides was normal. Cysteine specific activity was also measured afer [35S] cysteine injection and was found to be similar in treatment and control groups. The results suggest that cysteamine inhibits the synthesis of SRIF-14, SRIF-28, and OXT and stimulates that of AVP.

33/7/38 (Item 11 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

05113689 BIOSIS NO.: 000081071813
MAGNETIC RESONANCE SPECTROSCOPY OF WHEAT PROTEINS A MAGIC-ANGLE-SPINNING CARBON-13 NMR AND AN ESR SPIN LABEL STUDY
AUTHOR: MOONEN J H E; HEMMINGA M A; GRAVELAND A
AUTHOR ADDRESS: PFT INTERNATIONAL, P.O. BOX 18, 3300 AA ZWIJNDRECHT,, THE NETHERLANDS
JOURNAL: J CEREAL SCI 3 (4). 1985 (RECD. 1986). 319-328. 1985
FULL JOURNAL NAME: Journal of Cereal Science
CODEN: JCSCD

RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The freeze-dried wheat protein fractions, glutenin and gliadin, mixtures of high molecular weight (HMW) subunits of glutenin from flour, and gliadin-enriched and glutenin-enriched fractions from a gluten ball, were investigated by cross-polarization magic-angle-spinning 13C nuclear magnetic resonance spectroscopy. No characteristic differences in the molecular structure of these systems could be recognized in the spectra obtained. Sharp resonances in the aliphatic and aromatic spectral regions in the glutenin-enriched fraction, which have been described by other workers, were not observed: these may be artefacts arising from impurities present in the materials used. Isolated HMW subunits 2, 3, 10 and 11 were studied in solution using electron spin resonance after alkylation of cysteine residues with a maleimide spin label . Spectra obtained in 3 M urea were identical for all HMW subunits, with similar rotational correlation times (.tau.c) of 0.22 .+-. 0.02 ns. Spectra obtained in a less denaturing solvent (10 mM acetic acid) indicated that .tau.c increased for all HMW subunits ranging from 0.38 to 0.52 ns and that .tau.c for HMW subunit 10 was significantly lower than .tau.c for subunit 11. It is suggested that a high spin label mobility in 10 mM acetic acid is related to a better crosslinking function of subunit 10 in forming the extended, disulphide-bonded network of glutenin.

33/7/39 (Item 12 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

04285685 BIOSIS NO.: 000078015227
FACTORS AFFECTING THE LABELING OF HUMAN FIBRINOGEN WITH TECHNETIUM-99M IN-VITRO
AUTHOR: LAVIE E; SHENBERG C; BITTON M; MECHLIS S
AUTHOR ADDRESS: RADIOPHARM. DEP., SOREQ NUCL. RES. CENT., YAVNE 70600, ISRAEL.
JOURNAL: INT J APPL RADIAT ISOT 35 (2). 1984. 99-102. 1984
FULL JOURNAL NAME: International Journal of Applied Radiation and Isotopes
CODEN: IJARA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The preparation of [99mTc]fibrinogen is described. The following factors which could affect the quality of this preparation were studied: pH, SnCl2 and protein concentrations, pre- and post - labeling incubation times and labeling temperature. A 90% labeling yield was achieved by incubating fibrinogen with SnCl2 at pH 9.8 for 22 h before adding the TcO4- solution. The product obtained was stable and was 85% clottable. The X-ray fluorescence technique was employed to measure the amount of bound Sn at different times. [This labeled compound could play a key role in evaluating deep vein thrombosis in man.]

33/7/40 (Item 13 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

03331134 BIOSIS NO.: 000072059238

INFLUENCE OF CYSTEINE SPECIFIC LABELS ON THE STABILITY OF NUCLEOSOMES
AUTHOR: WINGENDER E; MAASS K; BODE J
AUTHOR ADDRESS: GESELLSCHAFT FUR BIOTECHNOL. FORSCHUNG, ABTEILUNG MOLEKULARBIOL., BRAUNSCHWEIG-STOECKHELM, W. GERMANY.
JOURNAL: INT J BIOL MACROMOL 3 (2). 1981. 114-120. 1981
FULL JOURNAL NAME: International Journal of Biological Macromolecules
CODEN: IJBMD
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The environments of the histone-H3 cysteines 110 are probed on 2 types of nucleosomes, N and N*, N, containing a DNA segment of 180 base pairs and the 4 core histones, was prepared by the direct degradation of chicken erythrocyte chromatin; the reactivity of its cysteines towards a fluorogenic reagent, BIPM N-[p-(2-benzimidazoly)phenyl]maleimide, is related to the ionic strength dependent liberation of H3. N* is a reconstituted N, to which a fluorescein-thiol label was reversibly attached; its accessibility towards DTT [dithiothreitol] occurs at much lower salt concentrations than the above process. Independent dissociation experiments and the stability towards protamines apparently substantiate that this difference is due to a labilization of the nucleosome. The processes previously derived from the observation of Cys-110 labels evidently occurred at salt or denaturant concentrations which were lower than for the native nucleosome. Fluorescence changes of the bound labels are not linked to the dissociation of H3, i.e., they are distinct from the thiol accessibilities which were derived kinetically.

33/7/41 (Item 14 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

03265128 BIOSIS NO.: 000071078239
A SIMPLE FLUORESCENT POST LABELING TECHNIQUE WITH O PHTHALALDEHYDE FOR THE ANALYSIS OF PROTEINS BY POLY ACRYLAMIDE GEL ELECTROPHORESIS
AUTHOR: YAJIMA T; ITOH T; MUTOH S; YUKI H
AUTHOR ADDRESS: SCHOOL OF PHARMACEUTICAL SCIENCE, TOHO UNIVERSITY, 2-2-1, MIYAMA, FUNABASHI, CHIBA, 274, JAPAN.
JOURNAL: CHEM PHARM BULL (TOKYO) 28 (12). 1980 (RECD. 1981). 3696-3698. 1980
FULL JOURNAL NAME: Chemical and Pharmaceutical Bulletin (Tokyo)
CODEN: CPBTA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: A new polyacrylamide gel electrophoretic system with post - labeling of proteins with o-phthaladehyde (OPT) was developed. In the present system, tris, glycine and ammonium persulfate were replaced by triethanolamine, L-hydroxyproline and sodium persulfate, respectively. Proteins were visualized by dipping the gels in the OPT reagent for several minutes immediately after electrophoresis, and the gels were then scanned in a fluorescence densitometer. Bovine serum albumin [50 mg] could be detected by this technique.

33/7/42 (Item 15 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

02633432 BIOSIS NO.: 000067021493
THE CYTOCHROME C OXIDASE EC-1.9.3.1 BINDING SITE ON CYTOCHROME C DIFFERENTIAL CHEMICAL MODIFICATION OF LYSINE RESIDUES IN FREE AND OXIDASE BOUND CYTOCHROME C
AUTHOR: RIEDER R; BOSSHARD H R
AUTHOR ADDRESS: BIOCHEM. INST., UNIV., CH-8028 ZURICH, SWITZ.
JOURNAL: J BIOL CHEM 253 (17). 1978 6045-6053. 1978
FULL JOURNAL NAME: Journal of Biological Chemistry
CODEN: JBCHA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Ferricytochrome c forms complexes with isolated [beef heart or baker's yeast] cytochrome c oxidase (ferrocytochrome c:oxygen oxidoreductase, EC 1.9.3.1) with stoichiometries of 1:1 and 2:1. These complexes were studied in order to localize the binding site for cytochrome c oxidase on the cytochrome c molecule. Ferricytochrome c, either free or bound to cytochrome c oxidase, was acetylated (or methylated) with a trace amount of [3H]acetic anhydride (or formaldehyde/KB3H4) under conditions where the degree of labeling was below 1 mol of 3H label /mol of cytochrome c. Subsequently , 3H-labeled cytochrome c was fully modified with excess of nonradioactive reagents and mixed with fully [14C]acetylated (or [14C]methylated) cytochrome c. Labeled peptides were isolated and the reactivity of the lysine residues was quantitated from 3H/14C ratios. When cytochrome c was bound to cytochrome c oxidase, lysine residue 13, and the groups of residues in positions 7-8, 72-73, and 86-88 became less reactive relative to the remaining 11 lysine residues of the molecule by factors of 1.5 to 5. The shielding effect of the oxidase was greatest toward residue 13. This information suggests a binding site for cytochrome c oxidase which spans over the top and part of the left-hand side of the cytochrome c molecule. (Conventional top and front view of the molecule according to the method of Mandel et al. The binding site involves part of the NH2-terminal helix at the top right of the heme crevice, lysine residue 13 above the exposed heme edge, the section around residues 86 to 88 at the top left of the heme evice and part of the left-hand surface of the molecule. The positively charged lysine residues demarcating the binding site have been conserved during evolution. A mechanism of electron transfer from heme c via the exposed heme edge (pyrrole ring II) to cytochrome c oxidase would be compatible with the present findings.

33/7/43 (Item 16 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

02102643 BIOSIS NO.: 000063017635
METABOLISM OF ACETATE CARBON-14 IN NORMAL AND OPAQUE-2 ZEA-MAYS ENDOSPERM DURING DEVELOPMENT
AUTHOR: GUPTA D N; LODHA M L; MEHTA S L
JOURNAL: PHYTOCHEMISTRY (OXF) 15 (9). 1976 1379-1382. 1976
FULL JOURNAL NAME: PHYTOCHEMISTRY (Oxford)
CODEN: PYTCA
RECORD TYPE: Abstract

ABSTRACT: Acetate-[2-14C]metabolism by developing normal and opaque-2 maize endosperms showed considerable differences in incorporation of label into organic acids, protein and free amino acids. Protein synthesizing efficiency was higher in opaque-2 endosperm 15 days after pollination but in normal at later stages of development. The differences in incorporation of label were more pronounced at early stages of endosperm development. Differences between the 2 endosperms occurred in the labeling of aspartate and glutamate in the free amino acids at 15 days post-pollination and in protein amino acids at 25 days post-pollination. Label and specific activity in protein lysine was higher in opaque-2 than in the normal.

33/7/44 (Item 1 from file: 73)
DIALOG(R)File 73:EMBASE

06327425 EMBASE No: 1995352579
Synthesis of two series of cysteine - specific spin labels containing a sup 1sup 5N substituted nitroxide
Wu G.
Department of Anesthesia, Massachusetts General Hospital, Harvard Medical School,Boston, MA 02114 United States
Journal of Labelled Compounds and Radiopharmaceuticals (J. LABEL. COMPD. RADIOPHARM.) (United Kingdom) 1995, 36/12 (1173-1181)
CODEN: JLCRD ISSN: 0362-4803
DOCUMENT TYPE: Journal; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH

Synthesis of two series of spin labels with a cysteine-reactive group, either maleimide or methanethiosulfonate, and a sup 1sup 5N substituted nitroxide separated at variable distance is reported. In the synthesis, the cysteine-reactive groups were first incorporated into the target molecule and the sup 1sup 5N substituted nitroxide was introduced in the last step via a mild reaction without destroying the cysteine-reactive groups. The synthetic routes described here are applicable to the synthesis of spin labels with either sup 1sup 5N substituted or 'normal' sup 1sup 4N nitroxide. Spin labeling of cysteine residues in rhodopsin by the newly synthesized spin labels was also demonstrated.

33/7/45 (Item 2 from file: 73)
DIALOG(R)File 73:EMBASE

03110219 EMBASE No: 1986222796
Effects of cysteamine administration on the in vivo incorporation of (sup 3sup 5S) cysteine into somatostatin-14, somatostatin-28, arginine vasopressin, and oxytocin in rat hypothalamus
Cameron J.L.; Fernstrom J.D.
Neuroendocrine Program, Department of Psychiatry, Western Psychiatric Institute and Clinic, Pittsburgh, PA 15213 United States
Endocrinology (ENDOCRINOLOGY) (United States) 1986, 119/3 (1292-1297)
CODEN: ENDOA
DOCUMENT TYPE: Journal
LANGUAGE: ENGLISH

The effect of cysteamine injection on the in vivo incorporation of (sup 3sup 5S) cysteine into somatostatin-14 (SRIF-14), SRIF-28, arginine vasopression (AVP), and oxytocin (OXT) in rat hypothalamus was studied. (sup 3sup 5S) Cysteine was injected into the third ventricle 1 h, 4 h, or 1 week after cysteamine (300 mg/kg, sc) injection; animals were killed 4 h later. The drug was found to substantially reduce immunoreactive SRIF levels, but not OXT or AVP, 4 h after its injection. Cysteamine also caused large reductions in label incorporation into SRIF-14, SRIF-28, and OXT 1 and 4 h after drug injection. However, (sup 3sup 5S) cysteine incorporation into AVP was increased substantially at these time points, while that into acid-precipitable protein was normal. One week after cysteamine injection, label incorporation into all hypothalamic peptides was normal. Cysteine specific activity was also measured after (sup 3sup 5S) cysteine injection and was found to be similar in treatment and control groups. The results suggest that cysteamine inhibits the syntheses of SRIF-14, SRIF-28, and OXT and stimulates that of AVP.

33/7/46 (Item 3 from file: 73)
DIALOG(R)File 73:EMBASE

00320754 EMBASE No: 1975093093
Intrinsic enzymatic activity of bovine procarboxypeptidase A S$D5
Uren J.R.; Neurath H.
Dept. Biochem., Univ. Washington, Seattle, Wash. 98195 United States
Biochemistry (BIOCHEMISTRY) 1974, 13/17 (3512-3520)
CODEN: BICHA
DOCUMENT TYPE: Journal
LANGUAGE: ENGLISH

Bovine procarboxypeptidase A S$D5 possesses inherent catalytic activity toward ester and peptide substrates of carboxypeptidase A. A kinetic analysis of this zymogen activity demonstrates that for ester substrates k(cat) is 5% that of carboxypeptidase A and K(m) is 100-300 times larger. For peptide substrates k(cat) is 1% to 4% that of the enzyme but K(m) is relatively unchanged. The inhibition constants (K(i)) for the dipeptide glycyl L tyrosine are also similar for the zymogen and the enzyme, suggesting that the substrate binding site is preformed in the zymogen. The catalytically functional elements of carboxypeptidase A (zinc, tyrosine, and glutamic acid) are also involved in the activity of the zymogen. Chelation of zinc with 1,10 phenanthroline inhibits both peptidase and esterase activity. Nitration of tyrosine with tetranitromethane abolishes peptidase activity and diminishes esterase activity. Following activation, peptidase activity remains negligibly low but esterase activity increases to 140% of that of native carboxypeptidase. Carboxyl modification with bromoacetyl N methyl L phenylalanine inhibits peptidase activity of the zymogen and prevents zymogen activation. When the zymogen is labeled with radioactive reagent and subsequently degraded, the radioactive label is found in a dipeptide (Phe Glu), which is identical with that obtained from similarly labeled carboxypeptidase. The pH dependence of carboxyl modification of the zymogen by Woodward's reagent K corresponds to the ionization of a group having a pK(a) of 5.0 as compared to 7.0 for the enzyme. It is proposed that activation of procarboxypeptidase A induces a change in conformation which alters the hydrogen bond structure of Gluinf 2inf 7inf 0 and enhances the reactivity of this nucleophile.

33/7/47 (Item 1 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

136211909 CA: 136(14)211909t PATENT
Human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant production and uses in inhibiting angiogenesis
INVENTOR(AUTHOR): Mazar, Andrew P.; Juarez, Jose C.
LOCATION: USA
ASSIGNEE: Attenuon, LLC
PATENT: PCT International ; WO 200214369 A2 DATE: 20020221
APPLICATION: WO 2001US23185 (20010724) *US PV220194 (20000724)
PAGES: 84 pp. CODEN: PIXXD2 LANGUAGE: English CLASS: C07K-014/81A; C07K-019/00B; C12N-015/62B; C12N-015/15B; C07K-016/38B; A61K-051/08B; A61K-038/57B; G01N-033/68B; C12N-005/08B; C12N-005/10B; A61K-047/48B
DESIGNATED COUNTRIES: AE; AG; AL; AM; AT; AU; AZ; BA; BB; BG; BR; BY; BZ; CA; CH; CN; CO; CR; CU; CZ; DE; DK; DM; DZ; EE; ES; FI; GB; GD; GE; GH; GM; HR; HU; ID; IL; IN; IS; JP; KE; KG; KP; KR; KZ; LC; LK; LR; LS; LT; LU; LV; MA; MD; MG; MK; MN; MW; MX; MZ; NO; NZ; PL; PT; RO; RU; SD; SE; SG; SI; SK; SL; TJ; TM; TR; TT; TZ; UA; UG; US; UZ; VN; YU; ZA; ZW; AM; AZ; BY; KG; KZ; MD; RU; TJ; TM DESIGNATED REGIONAL: GH; GM; KE; LS; MW; MZ; SD; SL; SZ; TZ ; UG; ZW; AT; BE; CH; CY; DE; DK; ES; FI; FR; GB; GR; IE; IT; LU; MC; NL; PT; SE; TR; BF; BJ; CF; CG; CI; CM; GA; GN; GQ; GW; ML; MR; NE; SN; TD; TG
SECTION:
CA203003 Biochemical Genetics
CA201XXX Pharmacology
CA206XXX General Biochemistry
CA213XXX Mammalian Biochemistry
IDENTIFIERS: sequence cDNA kininogen D5 domain human, angiogenesis inhibitor kininogen D5 domain human
DESCRIPTORS:
Ligands...
affinity, binding to; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Antiarteriosclerotics...
antiatherosclerotics; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Thioredoxins...
as binding partner; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Eukaryote(Eukaryotae)...
as host; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Allophycocyanins... Chemiluminescent substances... Chromophores... Color formers... Fluorescent substances... Phosphorescent substances... Phycocyanins... Phycoerythrins...
as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Nervous system...
ataxia telangiectasia, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Hyperplasia...
benign, treatment of; human kininogen D5 domain polypeptides, protein

and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Luminescent substances...
bioluminescent, as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Proteins...
calmodulin-binding, as binding partner; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Fibrosis...
chemotherapy-induced, assocd. with chronic inflammation, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Circulation...
coronary, collateral, disorder of, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Menstruation... Ovulation...
disorder of, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Pregnancy...
disorder, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Hematopoiesis...
disorders, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Protein motifs...
D5 domain; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Dopamine receptors...
D5, used in isolation of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Uterus,disease...
endometriosis, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Lung,disease...
fibrosis, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Anticoagulants...
for deep venous; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Angiogenesis inhibitors...
for myocardial or ischemic limb; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Bone,disease...
fracture, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Antitumor agents...

granulosa cell tumor, for pyogenic or neovascular; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Ovary,neoplasm...
granulosa cell tumor, inhibitors, for pyogenic or neovascular; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Immunoglobulins...
G1, fusion products; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Immunoglobulins...
heavy chains, hinge, CH2 or CH3, fused with kininogen D5 domain; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Angiogenesis inhibitors... Antiarthritics... Antibodies... Antidiabetic agents... Antitumor agents... cDNA sequences... Endothelium... Fusion proteins(chimeric proteins)... Human... Kininogens... Molecular cloning... Plasmid vectors... Protein sequences... Radionuclides... Signal transduction,biological... Virus vectors... Wound healing promoters...
human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Antibodies...
humanized; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Apoptosis...
inducing; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Cell proliferation...
inhibition, of endothelial cells; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Cell migration...
inhibition of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Drug delivery systems...
injections; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Spinal cord...
injury, scarring or fibrosis, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Artery,disease...
intima, hyperplasia, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Spinal cord...
ischemia, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Eye,disease...
keratopathy, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Antitumor agents...
leukemia; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Epitopes...
linear or conformational; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Antitumor agents...
lymphoma; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Eye,disease...
macula, degeneration, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Animal cell...
mammalian, as host; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Proteins...
MBP (maltose-binding protein), as binding partner; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Antitumor agents...
metastasis; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Diagnosis...
mol.; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Antibodies...
monoclonal; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Angiogenesis...
neovascularization, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Fluorescent substances...
Oregon Green, as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Antiulcer agents...
peptic; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Fibroblast...
proliferation, disorder of, retrolental, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Artery,disease...
restenosis, post-balloon angioplasty or vascular graft, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Eye,disease...
retinopathy, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Eye,disease...
rubeosis, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
Keloid...
treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

Blood vessel,disease...
vasculogenesis, treatment of; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
CAS REGISTRY NUMBERS:
402061-21-6P amino acid sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
50812-37-8 as binding partner; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
643-79-8 2321-07-5 13558-31-1 38183-12-9 82354-19-6 183185-51-5 189200-71-3 as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
10028-17-8 13981-27-6 14119-09-6 14133-76-7 14762-75-5 15064-65-0 15117-53-0 15715-08-9 15750-15-9 15755-33-6 15757-14-9 15758-35-7 biological studies, as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
10043-66-0 10098-91-6 14158-31-7 14391-96-9 14981-64-7 15092-94-1 15755-39-2 15757-86-5 29901-95-9 biological studies, human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
9001-90-5 9002-04-4 9004-08-4 9039-53-6 81669-70-7 cleavage of linker peptide by; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
2321-07-5D derivs., as label; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
268728-70-7 epitope H5-10 sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
268728-71-8 epitope H5-13 sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
268728-72-9 epitope H5-14 sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
401895-01-0 401895-02-1 epitope sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
605-65-2 for labeling; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
401895-03-2 401895-04-3 linker sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
402061-22-7 nucleotide sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
402061-33-0 402061-34-1 unclaimed nucleotide sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
402061-32-9 402061-35-2 unclaimed protein sequence; human kininogen D5 domain polypeptides, protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis
401895-05-4 unclaimed sequence; human kininogen D5 domain polypeptides,

protein and cDNA sequence, recombinant prodn. and uses in inhibiting angiogenesis

33/7/48 (Item 2 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

132352802 CA: 132(26)352802g PATENT
Post-labeling stabilization of radiolabeled proteins and peptides
INVENTOR(AUTHOR): Zamora, Paul O.; Merek, Michael J.
LOCATION: USA
ASSIGNEE: Rhomed Incorporated
PATENT: United States ; US 6066309 A DATE: 20000523
APPLICATION: US 794311 (19970203) *US PV11027 (19960202) *US 794270 (19970131)
PAGES: 21 pp., Cont.-in-part of U.S. Ser. No. 794,270, abandoned.
CODEN: USXXAM LANGUAGE: English CLASS: 424001490; A61K-051/00A; A61M-036/14B
SECTION:
CA263006 Pharmaceuticals
CA208XXX Radiation Biochemistry
CA209XXX Biochemical Methods
IDENTIFIERS: ascorbate stabilizer radiolabel protein peptide
DESCRIPTORS:
Diagnosis...
agents; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Imaging agents... Stabilizing agents... Test kits...
ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Drug delivery systems...
carriers; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Drug delivery systems...
freeze-dried; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Globulins,biological studies...
.gamma.-, technetium-99m derivs.; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Immunoglobulins...
M, anti-SSEA-1; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Antibodies...
monoclonal, anti-SSEA-1; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
Antigens...
SSEA-1 (stage-specific embryonic antigen 1), antibodies to; ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
CAS REGISTRY NUMBERS:
69-79-4 304-59-6 6381-92-6 23288-60-0 ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
50-81-7 56-40-6 110-15-6 biological studies, ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
14378-26-8DP cyclic somatostatin deriv., biological studies, ascorbate for post-labeling stabilization of radiolabeled proteins and peptides
7440-31-5 reactions, stannous ion; ascorbate for post-labeling

stabilization of radiolabeled proteins and peptides
103222-11-3DP rhenium-188 derivs., ascorbate for post-labeling
stabilization of radiolabeled proteins and peptides

33/7/49 (Item 3 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

127119073 CA: 127(9)119073g JOURNAL
Metal complexes with macrocyclic ligands. Part 44. Kinetics of the Cu2+ incorporation into a macrocyclic ligand conjugated to proteins. Model studies for the "post-labeling" technique
AUTHOR(S): Manzetti, Matthias; Macko, Ludwig; Neuburger-Zehnder, Margareta; Kaden, Thomas A.
LOCATION: Inst. Inorganic Chem., CH-4056, Basel, Switz.
JOURNAL: Helv. Chim. Acta DATE: 1997 VOLUME: 80 NUMBER: 3 PAGES: 934-947 CODEN: HCACAV ISSN: 0018-019X LANGUAGE: English PUBLISHER: Verlag Helvetica Chimica Acta
SECTION:
CA208009 Radiation Biochemistry
CA278XXX Inorganic Chemicals and Reactions
IDENTIFIERS: cyclam protein conjugate copper coordination kinetics, crystal structure copper cyclam complex
DESCRIPTORS:
Complexation kinetics...
kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins
Crystal structure... Molecular structure...
of (((tetraazacyclotetradecyl)methyl)benzoic acid)copper dichloride
Serum albumin...
4-((1,4,8,11-tetraazacyclotetradec-1-yl)methyl)benzoyl conjugate; kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins
CAS REGISTRY NUMBERS:
127925-35-3P crystal structure; kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins
556-53-6 10125-13-0 107288-23-3 127995-88-4 192637-25-5P kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins
15158-11-9 properties, kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins
9001-99-4DP 4-((1,4,8,11-tetraazacyclotetradec-1-yl)methyl)benzoyl conjugate, kinetics of copper(+2) coordination into macrocyclic ligand conjugated to proteins

33/7/50 (Item 4 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

122209130 CA: 122(17)209130z JOURNAL
A post-labeling technique for the iodination of DNA damage recognition proteins
AUTHOR(S): Billings, Paul C.; Cryer, Jonathan E.; Moy, Lily Y.; Engelsberg, Beatrice N.
LOCATION: School of Medicine, University of Pennsylvania, Philadelphia, PA, 19104, USA

JOURNAL: Cancer Biochem. Biophys. DATE: 1995 VOLUME: 14 NUMBER: 4
PAGES: 223-30 CODEN: CABCD4 ISSN: 0305-7232 LANGUAGE: English
SECTION:
CA209016 Biochemical Methods
CA201XXX Pharmacology
IDENTIFIERS: labeling iodination DNA damage recognition protein
DESCRIPTORS:
Iodination...
a post-labeling technique for iodination of DNA damage recognition proteins
Proteins,processes...
DNA damage recognition; a post-labeling technique for iodination of DNA damage recognition proteins
CAS REGISTRY NUMBERS:
15663-27-1 51592-06-4 a post-labeling technique for iodination of DNA damage recognition proteins

33/7/51 (Item 5 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

94204243 CA: 94(25)204243g JOURNAL
Influence of cysteine-specific labels upon the stability of nucleosomes
AUTHOR(S): Wingender, Edgar; Maass, Karin; Bode, Juergen
LOCATION: Abt. Molekularbiol., Ges. Biotechnol. Forsch., Braunschweig-Stoeckheim, Fed. Rep. Ger.
JOURNAL: Int. J. Biol. Macromol. DATE: 1981 VOLUME: 3 NUMBER: 2
PAGES: 114-20 CODEN: IJBMDR LANGUAGE: English
SECTION:
CA006013 General Biochemistry
IDENTIFIERS: nucleosome stability histone fluorescence label
DESCRIPTORS:
Nucleosome...
fluorescence labeling of cysteine of histone H3 of, stability in relation to
Histones,H3...
fluorescence labeling of cysteine of, nucleosome stability in relation to
Salt effect...
on nucleosome dissocn., fluorescence labeling of histone H3 in relation to
CAS REGISTRY NUMBERS:
52-90-4 biological studies, of histone H3, fluorescence labeling of, nucleosome stability in relation to
27030-97-3 cysteine of histone H3 labeling by, nucleosome stability in relation to

33/7/52 (Item 6 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

94099075 CA: 94(13)99075d JOURNAL
A simple fluorescent post-labeling technique with o-phthalaldehyde for the analysis of proteins by polyacrylamide gel electrophoresis
AUTHOR(S): Yajima, Takehiko; Ito, Toshihiko; Mutoh, Satoshi; Yuki,

Hidetaka
LOCATION: Sch. Pharm. Sci., Toho Univ., Chiba, Japan, 274
JOURNAL: Chem. Pharm. Bull. DATE: 1980 VOLUME: 28 NUMBER: 12 PAGES: 3696-8 CODEN: CPBTAL ISSN: 0009-2363 LANGUAGE: English
SECTION:
CA009003 Biochemical Methods
IDENTIFIERS: protein electrophoresis post labeling, phthalaldehyde albumin fluorescence electrophoresis
DESCRIPTORS:
Albumins,blood serum...
electrophoresis of, phthalaldehyde fluorescence staining after
Electrophoresis and Ionophoresis,gel...
of proteins, phthalaldehyde fluorescence staining after
CAS REGISTRY NUMBERS:
643-79-8 in protein fluorescence detn., after electrophoresis

RS1.C41

33/7/53 (Item 7 from file: 399)
DIALOG(R)File 399:CA SEARCH(R)

81147943 CA: 81(23)147943r JOURNAL
Sulfhydryl-specific fluorescent label, S-mercuric N-dansylcysteine. Titrations of glutathione and muscle proteins
AUTHOR(S): Leavis, Paul C.; Lehrer, Sherwin S.
LOCATION: Dep. Muscle Res., Boston Biomed. Res. Inst., Boston, Mass.
JOURNAL: Biochemistry DATE: 1974 VOLUME: 13 NUMBER: 15 PAGES: 3042-8
CODEN: BICHAW LANGUAGE: English
SECTION:
CA909004 Biochemical Methods
CA906XXX General Biochemistry
IDENTIFIERS: mercapto group detn reagent, protein labeling fluorescent reagent, mercuric dansylcysteine fluorescence protein
DESCRIPTORS:
Troponins...
C, fluorescence labeling of, with mercuric dansylcysteine
Mercapto group...
detn. of, by fluorescence, with mercuric dansylcysteine
Actins...
F- and G-, fluorescence labeling of, with mercuric dansylcysteine
Proteins... Tropomyosins...
fluorescence labeling of, with mercuric dansylcysteine
CAS REGISTRY NUMBERS:
53509-71-0 as fluorescent label, in mercapto group detn. and protein labeling
70-18-8 titrn. of, with mercuric dansylcysteine

33/7/54 (Item 1 from file: 351)
DIALOG(R)File 351:Derwent WPI

013628231
WPI Acc No: 2001-112439/200112
New composition comprising a carrier and a complex made from nucleic acid molecules and a charged copolymer useful in e.g. wound healing and for coating onto catheters is new

Patent Assignee: PLANK C (PLAN-I); SCHERER F (SCHE-I); STEMBERGER A (STEM-I)
Inventor: PLANK C; SCHERER F; STEMBERGER A
Number of Countries: 095 Number of Patents: 004
Patent Family:

Patent No	Kind	Date	Applicat No	Kind	Date	Week	
WO 200100708	A1	20010104	WO 2000EP5778	A	20000621	200112	B
AU 200052228	A	20010131	AU 200052228	A	20000621	200124	
DE 19956502	A1	20010531	DE 1056502	A	19991124	200131	
EP 1198489	A1	20020424	EP 2000936907	A	20000621	200235	
			WO 2000EP5778	A	20000621		

Priority Applications (No Type Date): DE 1056502 A 19991124; EP 99112260 A 19990625
Patent Details:

Patent No	Kind	Lan	Pg	Main IPC	Filing Notes
WO 200100708	A1	G	105	C08G-065/329	

Designated States (National): AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW
Designated States (Regional): AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW

AU 200052228	A		C08G-065/329	Based on patent WO 200100708
DE 19956502	A1		C12N-015/87	
EP 1198489	A1	G	C08G-065/329	Based on patent WO 200100708

Designated States (Regional): AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI

Abstract (Basic): WO 200100708 A1

NOVELTY - New gene therapy compositions comprise a carrier and a complex made from one or more nucleic acid molecules and a charged copolymer.

DETAILED DESCRIPTION - A composition of a carrier and a complex is claimed. The complex comprises one or more nucleic acid molecules and a charged copolymer of formula (R-W-(X-Zm-En)l-Y)p.

R=an amphiphilic polymer or a corresponding homo- or hetero-bifunctional derivative;

X=(1) an amino acid, peptide or spermine, or corresponding derivatives of these, (2) C(-a)(-b)(-c)(-d), (3) N(-a)(-b)(-c) or (4) an aromatic compound with three functional groups, W1, Y1 and Z1;

a=H or 1-6C alkyl (optionally substituted by halogen or dialkylamino);

b, c, d=1-6C alkylene (optionally substituted by halogen or dialkylamino);

W, Y, Z=CO, NH, O or S or a linker group which can react with SH, OH, NH or NH2;

E=an effector molecule which is a cationic or anionic peptide, spermine or spermidine (or corresponding derivatives of these), a glycosaminoglycan or a non-peptidic oligo/polycation or anion;

m, n=0-2;

p=3-20;

l=1-5.

ACTIVITY - Vulnerary.

MECHANISM OF ACTION - Gene therapy.

USE - The composition is useful for transfecting cells (claimed). The composition is useful for gene therapy. It can also be used as a

coating for implants (e.g. catheters) and to cover and help heal large areas of skin defect e.g. burns or decubitus ulcers, and as a carrier for tissue engineering.

ADVANTAGE - The charged copolymer coats the nucleic acid, providing improved stability and protection against opsonization.

pp; 105 DwgNo 0/22

Derwent Class: A96; B04; D16

International Patent Class (Main): C08G-065/329; C12N-015/87

International Patent Class (Additional): A61K-047/48; A61K-048/00; C08G-065/333

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